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MICROHISTOLOGICAL CHARACTERISTICS OF SELECTED AQUATIC PLANTS OF FLORIDA

with Techniques for the Study of Manatee Food Habits



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MICROHISTOLOGICAL CHARACTERISTICS OF SELECTED AQUATIC PLANTS OF FLORIDA

with Techniques for the Study of Manatee Food Habits

by

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PREFACE

Studies of terrestrial mammalian herbivore diets are numerous, whereas little information exists on the food habits of aquatic mammalian herbivores. Previous to this report there have been no published techniques for the analyses of ingesta of Florida manatees (*Trichechus manatus latirostris*) for identification of dietary components. The lack of previous work necessitated research and development of techniques for food item identification and quantification in order to provide a standardized method for the study of the manatee diet.

Descriptions of the microscopic characters of the plants featured in this report are based on morphology; function is not considered. Terminology used is descriptive and the authors' descriptions of gross shape under low (<200X) magnification are designed to facilitate practical classification of unknown epidermal features by a technician with little detailed knowledge of plant anatomy. Nevertheless, some intergrading forms of microanatomical characters remain difficult to categorize. For example, the distinction between elongated prickles and short, robust macrohairs; numerous thin styloids and fragmented raphides; or any of the silica body shapes is qualitative and often subjective.

As a reference manual this report is useful only with respect to the taxa listed in the index. The material contained here will require revision as new species are observed or suspected in the manatee diet. It must be emphasized that the data on which this study is based are not comprehensive. Even so, this study should guide and encourage identification of fragmented material in food habits analyses of the diet of manatees in Florida.

Use of this report for food habits studies of manatees or other aquatic herbivores in other localities will require additional work in collecting, identifying, preparing and observing potential food items of the local area. Indeed, additional material should be incorporated continually into the collection from areas of Florida not yet represented.

Questions or comments regarding this report should be directed to:

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INTRODUCTION

The fragile nature of aquatic and wetland plants, coupled with the effects of mastication and digestion, requires the use of microhistological analysis to identify food fragments in the ingesta or feces of the manatee. Microhistological analysis of the herbivore diet must begin with identification and cataloguing of plants in the herbarium as well as in a microscope slide collection. These specimens then can be classified into distinct groups on the basis of microanatomical features to allow identification of the small fragments found in manatee ingesta. A key must be developed using these important microscopic features to identify and analyze unknown ingesta fragments.

This study was initiated in 1978 to develop a technique of identifying and quantifying the digestive tract contents of Florida manatees (*Trichechus manatus latirostris*) and to serve as a manual for the identification and analysis of ingesta collected from manatee carcasses salvaged in Florida.

This report includes key microhistological characters found useful in identifying fragments of 83 plant species and three invertebrate groups. Many species of aquatic and wetland plants and invertebrates are available to manatees in Florida as potential foods. The taxa chosen for study in this report were selected on the basis of their occurrence in habitats known to be utilized by manatees as well as on the basis of their observed, reported, or suspected consumption by manatees. Our objectives were to provide: (1) a description and illustration of important microhistological features of selected potential manatee foods; (2) a provisional key to enable workers to identify plant and invertebrate fragments salvaged from manatee ingesta or fecal samples; (3) recommended methods and materials for the quantitative analysis of manatee ingesta; and (4) a starting point for further work on microanatomical identification of the aquatic and wetland plants found in manatee habitats in Florida.

METHODS

IDENTIFICATION OF FOOD ITEMS IN MANATEE INGESTA

A collection of several hundred specimens representing nearly 100 species of selected plants and invertebrates was compiled by the U.S. Fish and Wildlife Service Sirenia Project personnel and identified by appropriate authorities. The plant specimens were collected mostly in Florida, in areas frequented by manatees.

Microhistological analysis of manatee ingesta and fecal material required reference plant and invertebrate specimens to be mounted on slides for easy study. Reference or voucher specimens of potential food items were collected and preserved with appropriate field data for later study in their entirety (preserved dry or wet) and on microscope slides. Slide reference material included the portion of the organism eaten most often, such as the leaf, stem, root, or flower. According to the limited studies of stomach and fecal samples from manatees in Florida, leaf and stem material were apparently the most frequently eaten parts of plants.

Leaf (and sometimes flower, seed, stem, rhizome, and root) material from the identified and catalogued voucher specimens was permanently mounted on slides and organized by taxon according to type (algae, seagrasses, freshwater aquatics, grasses, and forbs, shrubs, and trees). Photomicrographs of these reference slides were taken at 20X, 50X, and 100X magnifications and assembled into a 10-volume photocatalogue to facilitate comparison of each taxon's epidermal characteristics. A diagnostic key, character matrix tables of obvious histological features, and summary tables of key characters, were constructed to aid in the identification of unknown plant fragments recovered from manatee ingesta.

References useful in the development of the methods and key included Carter and Hestand (1979) and Johnson et al. (1983). Identification of plants was assisted using Godfrey and Wooten (1979, 1981), Tarver et al. (1979), Taylor (1960), and Whitford and Schumacher (1973).

The anatomical work of Metcalfe (1960) on monocotyledons and Metcalfe and Chalk (1950) on dicotyledons provided the general guidelines for the study of microhistological characters used in this project. Some of the features found to be important and useful for microscopic analysis were: epidermal cell shape, arrangement; stoma type, size, and arrangement; characteristics; trichome type, size, and placement; presence and shape of idioblasts; crystal type and size; and silica body shape and arrangement. Leaf size and the presence of tannin bodies were of less importance because of the high variability in these characters. Although dimensions of microhistological characters are more liable to vary with environment than differences in kind (Metcalfe and Chalk 1950), lengths and widths of many characters were included in this report to provide additional identifying information. Seldom did one singular microanatomical characteristic provide for accurate identification. Rather, a combination of features, using both qualitative and quantitative criteria, was necessary and most effective in determining the identity of an unknown fragment.

REFERENCE SLIDE PREPARATION

Fresh plant material (leaf and stem) was mounted on microscope slides for observation under 100X magnification. The material often was boiled in Hertwig's solution to facilitate clearing of epidermal material. Scraping the leaf or stem with a scalpel until only the epidermis of one side remained was necessary if the plant had thick leaves or stems. Scraping also was advantageous with thin-leaved plants as it removed opposing epidermal cells that otherwise remained

apparent and confused observations of cell shapes and sizes. Epidermal scraping, however, had to be done with care as the process could remove veins, crystals, idioblasts, and other elements of the leaf mesophyll that were important features for microscopic identification. For this reason, plant material prepared for a reference slide collection was mounted both scraped and unscraped. Unscraped material was mounted whole and fragmented, as both of these mounts offered unique views of the available microhistological characters. Staining plant material was not necessary, and in fact was found to be detrimental, as tannin and other natural pigments often were obscured.

Specimen material was mounted in either Permount (Fisher Scientific Co.), Histoclad (Clay Adams Co.), or Hoyer's solution. Permount and Histoclad required soaking the specimen in alcohol or xylene to remove free water. The chief apparent difference between the two media was that Histoclad dried more quickly than Permount. Histoclad had the advantage of drying in 24 hours at room temperature and being impervious to humidity. Hoyer's solution is water soluble and required no special preparation for mounting the specimen. Hoyer's solution is hygroscopic, however, and had to be stored in a low humidity environment to prevent misalignment of the specimen. We preferred Hoyer's solution over Histoclad or Permount because it did not require the specimen to be dehydrated for mounting.

Regardless of which mounting medium was used, slide preparation methods for ingesta analysis had to be consistent with those used for voucher slide specimens. One example of an effect of inconsistent preparation methods (immersion of plant material in cold water versus immersion in hot water or alcohol) was differential precipitation of crystals in a grass leaf (Davies 1959).

The prepared slides were allowed to dry before observing under the microscope. Heating slides in an oven hastened drying, but temperatures higher than 60 °C caused bubbles to form under the cover slip. Dried slides prepared with Hoyer's solution were stored flat to prevent slipping of the material.

PHOTOGRAPHY

Photomicrographs of plant material were taken through a microscope with standard bright field illumination or polarized light, using 35mm Kodacolor print film (ASA 100); 50X magnification (10X objective and 5X camera adapter) of the material was most useful for later identification. Development of photographic prints increased magnification to about 205X. Unpolarized light required an exposure time of one second at 50X magnification, while polarized light required 10 seconds exposure time at the same magnification. Results were variable due to differences in thickness and color of specimens. A polarizing filter was useful in locating, identifying, and photographing crystals, idioblasts, spines, and vascular tissue in plant material, as these structures often were highly birefringent and gave high contrast in polarized light.

PREPARATION OF THE DIET SAMPLE

Manatee diet samples were collected from the field (fecal material) and from salvaged carcasses (ingesta). Samples were preserved and stored wet (in 5% NB formalin or 70% ethyl alcohol) or dried.

For analysis in the laboratory, preserved or fresh ingesta from the stomachs of salvaged specimens was preferred, due to their relatively larger fragment size. The samples were rinsed with tap water over a 30-mesh (0.52-mm) screen to remove dirt and other fine particles that hampered observation under the microscope. A small amount of washed ingesta was then spread evenly over a slide and observed at 100%. Food material was identified by comparing the ingesta fragments with the voucher microscope slides, photomicrographs, or illustrations in the reference collection.

RESULTS

Eighty-six taxa, representing freshwater and marine plants (34), algae (19), grasses and allies (17), forbs, shrubs, and trees (13), and invertebrates (3), were studied.

Important features observed in the taxa examined are illustrated in Figures 1-7. These characters were used often in the identification of microscopic fragments found in manatee ingesta. Profiles of each species, including descriptions of characteristic features and illustrations drawn from photomicrographs, are illustrated in Figures 8-90, at about 133X magnification, unless otherwise noted on the figure.

Distinct features and measurements of selected elements for all but one species (unidentified sponge) are summarized in Tables 1-4. Microscopic characters observed in most taxa and the species in which these were observed are listed in Tables 5-6.

Equipment and materials needed for microanalysis of manatee food habits are listed in Appendix A. The recommended method for analyzing manatee ingesta quantitatively is in Appendix B. Appendix B also includes samples of recording sheets used during examination of ingesta and fecal samples. A provisional diagnostic key for the 86 taxa, provided in Appendix C, was formulated using the most distinct microscopic features observed in each species. A taxon was incorporated in the key more than once when several combinations of characters were useful in distinguishing the taxons from each other. However, all characters are not always included for each set of choices. Therefore, it may be necessary to try more than one obvious character to identify a fragment.

In addition to this report, all voucher specimens were accessioned into a herbarium and on microscopic slides, and a catalogue of photomicrographs has been provided to the U.S. Fish and Wildlife Service, National Ecology Research Center, Sirenia Project laboratory. These materials were useful for comparative illustrations and reference.

Table 1. Character matrix of observed features for selected Florida forbs, shrubs, and trees.

SPECIES	-	2	ო	4	ည	9	7	ω	6	10	11	12
Carya glabra ANO Colocasia esculenta 3 Commelina diffusa * X Crinum americanum 4 X Diospyros virginiana ANO X Erechtites hieracifolia *ANO/ANI*X Fraxinus caroliniana ANO Magnolia grandiflora ANO Myrica cerifera ANO X Phyla nodiflora ANO X Rhizophora mangle * * * Ulmus americana ANO X	ANO 3 4 4 ANO/ANI ANO PAR ANO		4,6 6 3 * 2,6 1,2 1,2,4 1,2,4,6 - 1,2,3,6	0/CS D/R R/CS CS CS - 0/CS D/CS		:××××::::::×:	×××××××××××	4444-4-A4AA-	.015080 .010065 .015090 .0424 .0105 .0512 .015090 .0208 .0104 .0107 .0105	.0104 .0105 .0106 .0104 .0103 .015070 .0107 .0102 .0103 .0103	.015020 .035040 .0204 .0203 .035045 .015030 .020025 .015025 .020025	.010015 * .0204 .015030 .010010 .0203 .010015 .0102 .05015 .0102 .05015

Key to observed features: "X" = feature observed; "*" = feature not always present or distinguishable; "-" = feature not observed.

6 = Tannin cells observed 7 = Cross veins observed	8 = Epidermal cell shapes: A=angular; L=lobed; A/L=both	<pre>9 = Epidermal cell lengths (mm) 10 = Epidermal cell widths (mm)</pre>	<pre>11 = Stoma lengths (mm) 12 = Stoma widths (mm)</pre>	
<pre>1 = Stoma types: monocots: types 1,2,3, or 4;</pre>	<pre>dicots: ANI=anisocytic, ANO=anomocytic, PAR=paracytic 2 = Leaf edge spines/trichomes observed</pre>	<pre>3 = Trichome types: 1=globular; 2=hair; 3=prickle; 4=peltate; 5=stellate; 6=papillae</pre>	4 = Crystal types: CS=crystal sand: D=druse: R=raphide	5 = Idioblasts observed

Character matrix of observed features for selected Florida freshwater and marine plants. Table 2.

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Table 2. (Concluded)

SPECIES	-	2	ო	4	2	9	7	∞	6 .	10	11	12	13	14
Sagittaria subulata Salvinia rotundifolia Syringodium filiforme Thalassia testudinum Utricularia sp. Vallisneria americana Zannichellia palustris			. *	\$3. \$3	111111	× · ×× · ××	×× ·× ·××	* : :× :* :	A	пооооп	.0310 .0309 .010015 .01505 .015070	.0105 .0105 .005010 .0103 .0103		1 1 1 1 1 1 1

Key to observed features: " X " = feature observed; " * " = feature not always present or distinguishable; "-" = feature not observed

1 = Stoma types:	6 = Tannin cells observed
Monocots: 1,2,3 or 4;	7 = Cross veins observed
Dicots: anomocytic or paracytic	8 = Veins glow under polarized light
<pre>2 = Leaf edge spines/trichomes observed</pre>	9 = Epidermal cell shapes:
3 = Trichome types:	A = angular; L = lobed; A/L = both
1 = globular; 2 = hair; 3 = prickle;	10 = Leaf edge cell length relative to adjacent
4 = peltate; 5 = stellate; 6 = papillae	epidermal cells: S = short, L = long, E = equal
4 = Crystal types:	<pre>11 = Epidermal cell lengths (mm)</pre>
<pre>CS = crystal sand; D = druse; R = raphide;</pre>	12 = Epidermal cell widths (mm)
S = styloid; U = unidentified	13 = Stoma lengths (mm)
5 = Idioblasts observed	14 = Stoma widths (mm)

Character matrix of observed features for selected Florida grasses and allies. Table 3.

18	2-4	2	1-3	2	× × ×	2-4	.025 2	2-3	2	1-2	2	2	<u>۸</u>	2-4
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16	.0304	.0508	, -	.0818	.0308	.0408	.0408	ı	ı	ı	.04	.0410	.0610	.0306
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14	08/+	0/B	DB/N	DB/N	DB/+ -	DB/TN	DB/N	DB/+	DB/N/+	DB/N	DB/N/I	N/I	0/B	N/DB
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1	.020035 .0102	.015020	.025030	.025040		.0104	.0204	.0203	.030045	.0204	.0203	.0204	.0304	.035050 .015030
SPECIES	Brachiaria mutica	Spicata Spicata	muricata Febinochlos	paludigena Hudrochlos	caroliniensis.020025 Juncus sp0203	hexandra	Setarius Setarius Denicum	Commutatum Danicum	dichotomum	hemitomon Decre	rasparum distichum Decnelum	rasparum repens Dhusquitoc	australis Sacciologis	striata

(Continued)

(Concluded) Table 3.

SPECIES	н	2	က	4	2	9	7	8	9 10	3 4 5 6 7 8 9 10 11 12 13 14 15	12	13	14	15		16	17	18
Spartina alterniflora .015030 .010 X - X Typha domingensis .015030 .010015 - CS/R -	15030 .0	010	× ;	- X CS/R -	× '	1 1	1 1	× ,	× : × :	G 7	× '	1-3	0/E	LD X 1-3 0/B 0/I P		.0406 .05	.05	2 *
Key to observed features: " X " =	atures: "		feat	ture	opş	er.	/ed;	=	=	feature observed; " - " = feature not observed	ture	not	epse	rved				
<pre>1 = Stoma lengths (mm) 2 = Stoma widths (mm) 3 = Leaf edge spines 4 = Crystal types: CS=crystal sand; R=raphide 5 = Papillae 6 = Macrohairs 7 = Microhairs 8 = Interstomal cells with concave ends 9 = Prickles between veins</pre>	(mm) ines ines s: CS=crys cells with	tal san concav	d; -	R=ra nds	phic	ē			111 122 133 144 151 161 171		sidi low rt c ber ica ica ckle ckle	ary dome ells of r body body len	cell; P=k; pair pair shap shap shap shap gths	shape arall ed be for shown in the over the better (mm) for shown in the shown in the shown in the shape are shape are shown in the shape are shape	Subsidiary cell shapes: LD=low dome; P=parallel; T=triangular Short cells paired between veins Number of rows of short cells over veins Silica body shape over veins:* Silica body shape between veins:* Prickle lengths (mm) over veins Number of rows of stomata between veins	iangul ins over * ns:* ns veins	ar veins eins	

* Silica body shapes: B = block; + = cross; DB = dog bone; I = irregular; N = nodular; O = oval; TN = tall and narrow

8 = Interstomal cells with concave ends
9 = Prickles between veins
10 = Prickles over veins

Character matrix of observed features for selected Florida algae and invertebrates. Table 4.

SPECIES	1	2	က	4	က	ဖ	_	ω	6	10	11	12	13	14
Acanthophora spicifera Bugala neritina	× ·	×*	ı *	·×	A/L	· ×	1 1	1 1	* ×	1 1	××	.0312	.005030	ပပ
Caloglossa leprieurii Caulerpa prolifera	´ I I	× 1	ı ×	1 1	۲ A	1 1	1 1	· ×	1 1	1 1	××	.0320	.0105	O I
Chaetomorpha brachygona Chara Zeylanica Enteromorpha	· ×	3 I	××	1 1	4 1	× ·	· *	1 1	1 1	· ×	× ·	.245 0	.0314	SS
compressa Enteromorpha	1	1	×	4	Ø	1	×	1	•	ı	×	.0105	.008025	ပ
intestinalis	1	1	×	•	⋖	1	×		1	•	×	.0104	.008020	ပ
cervicornis Gracilaria	ı	×	1	1	_	ı	,	ı	ı	ı	×	.008020	.008020	ပ
verrucosa Hydroid	1 1	×ı	1 1	ı ×	. _ &	· ×	1 1	1 1	1 1	1 1	××	* *	* .0526	1 0
Hypnea cervicornis Jania adherens	1 1	××		1 1	4 4	××	* *	1 1	1 1	1 1	××	.1020	202	S
Oedogonium sp. Oscillatoria tenuis Polysiphonia	1 1	: 1 1	× 1	· ×	4 4	××		1 1	ı	××		· ·	ഥഥ	o vo
subtilissima Dterocladia	1	×	1	1	V	×	ι,	1	1	ı	×	.1020	.0407	S
americana Sardassum	1	×	1	ı	A	ı	•	ı		1	×	.00250100	.00250100	ပ
filipendula Soliera tenera	1 1	· ×	1 1	× 1		1 1	1 1	1 1	1 1	1 1	××	.015035	.0102 .001012	ပပ

(Continued)

Table 4. (Concluded)

SPECIES	1	2	3 4	4	2	9	7	80	5 6 7 8 9 10 11	10	11	12	13	14
Spirogyra sp. Ulva lactuca	1 1	1 1	××	1 1	A A	× ·		1 1	1 1	× ·	'×	.010025	.0314	νυ
Key to observed features: "X" = feature observed; "*" = feature not always present or distinguishable; "-" = feature not observed	"X" I Kan to a to	= fe = an fil ked illa	gula ame "A" le" (ar; ntous	Serve	d; 8 8 9 10 111 111 111 113 113 114 114	1; 8 8 1 10 10 11 11 11 11 11 11 11 11 11 11 1	= fe Idio Structuresh Tresh Tarin	"Idioblasts" observed; " structures resembling i Filaments glow under pol Freshwater habitat Marine habitat; ranking categories is based on t Whitford and Schumacher Cell/segment lengths (mm Cell/segment widths (mm) Measured feature: C = ce	not in ok il ok il ok habi it at; is b it le it le it wi	alwa alwa sembl unde itat ran sased shuma ungths	<pre>; "*" = feature not always present or distinguishable; 8 = "Idioblasts" observed; "idioblasts" on algae are structures resembling idioblasts on vascular plants 9 = Filaments glow under polarized light 10 = Freshwater habitat 11 = Marine habitat; ranking of algae into freshwater/marine categories is based on the literature (Taylor 1960; Whitford and Schumacher 1973) 12 = Cell/segment lengths (mm) 13 = Cell/segment widths (mm) 14 = Measured feature: C = cell; S = segment</pre>	distinguish ts" on algae s on vascula ight ature (Taylo segment	able; are r plants ater/marine r 1960;

Table 5. Characteristic features observed in grasses (Family Gramineae) and allies.

-	-	•		•	_	
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1 7		1	L.		E.,	•

Brachiaria mutica
Distichlis spicata
Echinochloa muricata
Echinochloa paludigena
Hydrochloa caroliniensis

Leersia hexandra Paspalum distichum Paspalum repens Spartina alterniflora

MACROHAIRS:

Hydrochloa caroliniensis Oplismenus setarius Paspalum repens

MICROHAIRS:

Brachiaria mutica
Echinochloa muricata
Echinochloa paludigena
Hydrochloa caroliniensis
Leersia hexandra
Oplismenus setarius

Panicum commutatum

Panicum dichotomum
Panicum hemitomon
Paspalum distichum
Paspalum repens
Phragmites australis
Sacciolepis striata

PRICKLES:

Brachiaria mutica
Distichlis spicata
Echinochloa muricata
Echinochloa paludigena
Hydrochloa caroliniensis
Leersia hexandra

Panicum dichotomum
Paspalum distichum
Paspalum repens
Phragmites australis
Sacciolepis striata
Spartina alterniflora

Panicum commutatum

LEAF EDGE SPINES:

Brachiaria mutica Echinochloa muricata Echinochloa paludigena Hydrochloa caroliniensis

Leersia hexandra Oplismenus setarius Panicum commutatum

Oplismenus setarius

Panicum dichotomum
Panicum hemitomon
Paspalum repens
Phragmites australis
Sacciolepis striata
Spartina alterniflora

CRYSTALS:

Brachiaria mutica - crystal sand

Hydrochloa caroliniensis - crystal sand

Sacciolepis striata - crystal sand

Typha domingensis - crystal sand, raphides

(Continued)

Table 5. (Continued)

SILICA BODY SHAPE OVER VEINS

BLOCK:

Distichlis spicata

Phragmites australis

CROSS:

Brachiaria mutica Panicum commutatum Panicum dichotomum

DOGBONE:

Brachiaria mutica
Echinochloa muricata
Echinochloa paludigena
Hydrochloa caroliniensis
Leersia hexandra

Panicum dichotomum Panicum hemitomon Paspalum distichum Sacciolepis striata

Panicum commutatum

Oplismenus setarius

IRREGULAR:

Paspalum distichum

Paspalum repens

NODULAR:

Echinochloa muricata Echinochloa paludigena Oplismenus setarius Panicum dichotomum

Panicum hemitomon Paspalum distichum Paspalum repens Sacciolepis striata

OVAL:

Distichlis spicata

Phragmites australis

TALL AND NARROW:

Leersia hexandra

SILICA BODY SHAPE BETWEEN VEINS

BLOCK:

Brachiaria mutica Distichlis spicata Echinochloa muricata Phragmites australis

(Continued)

Table 5. (Concluded)

CROSS:

Echinochloa muricata

Hydrochloa caroliniensis

Oplismenus setarius

DOG BONE:

Echinochloa muricata

Echinochloa paludigena

IRREGULAR:

Brachiaria mutica

Panicum dichotomum

Panicum hemitomon Spartina alterniflora

OVAL:

Distichlis spicata Panicum hemitomon

Paspalum distichum

Phragmites australis Spartina alterniflora

TALL AND NARROW:

Brachiaria mutica

Distichlis spicata Echinochloa muricata

Panicum hemitomon Paspalum distichum

Table 6. Characteristic features observed in aquatic plants, forbs, shrubs, and trees.

		AQUATICS	FORBS, SHRUBS AND TREES	
STOMATA:	ANO 3 PAR ANO ANO ANO 3	Alternanthera philoxeroides Eichhornia crassipes Hydrocotyle sp. Nuphar luteum Nymphaea mexicana Polygonum punctatum Sagittaria lancifolia Sagittaria stagnorum	Carya glabra Colocasia esculenta Commelina diffusa Crinum americanum Diospyros virginiana Erechtites hieracifolia Fraxinus caroliniana Magnolia grandiflora Myrica cerifera Phyla nodiflora Quercus nigra Rhizophora mangle Ulmus americana	ANO 3 * 4 ANO *ANO/ANI* ANO PAR ANO * ANO * ANO
LEAF EDGE SPINES OR TRICHOMES	•	Alternanthera philoxeroides Cabomba caroliniana Ceratophyllum demersum Egeria densa Halophila engelmannii Halophila sp. Hydrilla verticillata Najas guadalupensis Polygonum punctatum Potamogeton illinoensis Ruppia maritima Sagittaria kurziana Thalassia testudinum Utricularia sp. Vallisneria americana	Commelina diffusa Crinum americanum Diospyros virginiana Erechtites hieracifolia Myrica cerifera Rhizophora mangle Ulmus americana	*
TRICHOMES	7 7 7 * 1,6 1 1 1 2 1,2	Alternanthera philoxeroides Cabomba caroliniana Halophila sp. Hydrilla verticillata Hydrocotyle sp. Myriophyllum aquaticum Myriophyllum spicatum Nuphar luteum Nymphaea mexicana Pistia stratiotes Polygonum punctatum Sagittaria kurziana	Carya glabra Commelina diffusa Crinum americanum Diospyros virginiana Erechtites hieracifolia Fraxinus caroliniana Magnolia grandiflora Myrica cerifera Phyla nodiflora Rhizophora mangle Ulmus americana	4,6 2,3 * 2,6 1,2 1,2,4 1,2 1,2,4,6 2 * 1,2,3,6

(Continued)

Table 6. (Continued)

AQUATICS FORBS, SHRUBS AND TREES Sagittaria stagnorum Salvinia rotundifolia 2* Vallisneria americana D,CS Carya glabra CRYSTALS: Alternanthera philoxeroides Ceratophyllum demersum Colocasia esculenta D,R U Commelina diffusa R,CS R,S Eichhornia crassipes Halophila engelmannii Crinum americanum CS R Diospyros virginiana CS CS Halophila sp. Fraxinus caroliniana CS S*.R*.CS* Hydrocotyle sp. Myrica cerifera D.CS Lemna sp. R,D CS.D Quercus nigra CS,D,R,S Ludwigia repens Rhizophora mangle Myriophyllum aquaticum D D CS Ulmus americana D Myriophyllum spicatum U* Nuphar luteum U* Nymphaea mexicana D,R Pistia stratiotes Polygonum punctatum D U Sagittaria lancifolia CS* Sagittaria subulata Salvinia rotundifolia CS CS* Thalassia testudinum Colocasia esculenta Alternanthera philoxeroides TANNIN CELLS: Commelina diffusa Azolla caroliniana Cabomba caroliniana Crinum americanum Diospyros virginiana Ceratophyllum demersum Rhizophora mangle Egeria densa Eichhornia crassipes Halodule sp. Halophila engelmannii Halophila sp. Hydrilla verticillata ·*Hydrocotyle* sp. Ludwigia repens Myriophyllum aquaticum Myriophyllum spicatum Najas quadalupensis

(Continued)

Pistia stratiotes Ruppia maritima

AQUATICS

FORBS, SHRUBS AND TREES

Sagittaria kurziana Sagittaria lancifolia Sagittaria stagnorum Sagittaria subulata Syringodium filiforme Thalassia testudinum Vallisneria americana Zannichellia palustris

VEINS GLOW UNDER POLARIZED LIGHT: Egeria densa Eichhornia crassipes Halodule sp. Halophila engelmannii Hydrilla verticillata Najas guadalupensis Nuphar luteum Nymphaea mexicana Potamogeton illinoensis Potamogeton pectinatus Potamogeton pusillus Sagittaria kurziana Sagittaria lancifolia Sagittaria stagnorum Sagittaria subulata Thalassia testudinum Vallisneria americana

Commelina diffusa Fraxinus caroliniana Quercus nigra Rhizophora mangle

IDIOBLASTS:

Nuphar luteum Nymphaea mexicana Rhizophora mangle

CROSS VEINS:

Azolla caroliniana
Eichhornia crassipes
Halophila engelmannii
Halophila sp.
Hydrocotyle sp.
Lemna sp.
Ludwigia repens
Nuphar luteum
Nymphaea mexicana
Pistia stratiotes
Polygonum punctatum
Potamogeton illinoensis

Alternanthera philoxeroides

Carya glabra
Colocasia esculenta
Commelina diffusa
Crinum americanum
Diospyros virginiana
Erechtites hieracifolia
Fraxinus caroliniana
Magnolia grandiflora
Myrica cerifera
Phyla nodiflora
Quercus nigra
Rhizophora mangle
Ulmus americana

(Continued)

Table 6. (Concluded)

AQUATICS

FORBS, SHRUBS AND TREES

CROSS VEINS:

Potamogeton pectinatus Potamogeton pusillus Sagittaria kurziana Sagittaria lancifolia Sagittaria stagnorum Sagittaria subulata Salvinia rotundifolia Thalassia testudinum Vallisneria americana Zannichellia palustris

^{*}see Tables 1-3 for definitions of feature abbreviations

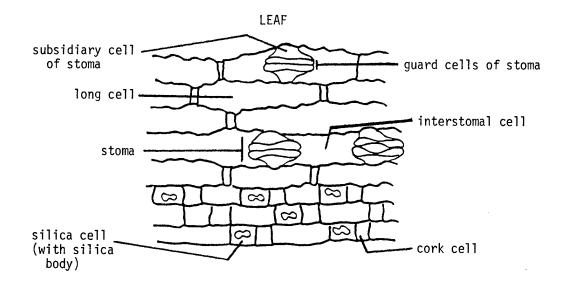
^{*}feature is not always present or distinguishable

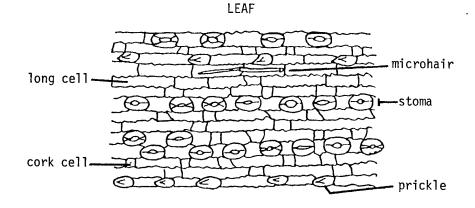
ANGULAR Brick-like Filamentous LOBED Sinuous Sinuous Anticlinal

Figure 1. Characteristic shapes of epidermal cells of selected species included in this report.

Irregularly Lobed

Round





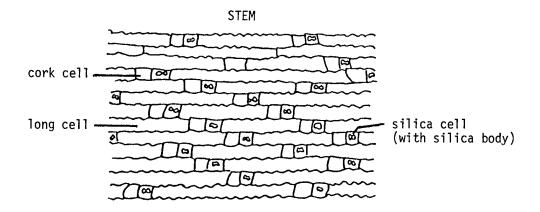
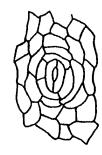


Figure 2. Epidermal characters of leaves and stems of selected species of grasses included in this report. (Adapted from Esau 1965 and Fahn 1974).

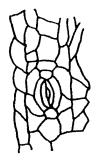
Type 1

The guard cells are surrounded by four to six subsidiary cells



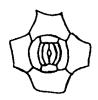
Type 2

The guard cells are surrounded by four to six subsidiary cells of which two are roundish, smaller than the rest, and are situated at the ends of the guard cells



Type 3

The guard cells are accompanied laterally by two subsidiary cells, one on each side



Type 4

The guard cells are not associated with any subsidiary cells

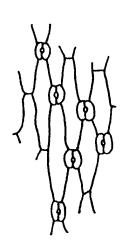
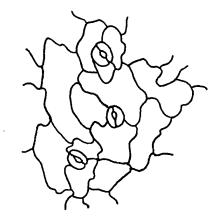


Figure 3. Stomatal arrangement in the leaf epidermis of monocotyledons included in this report (from Fahn 1974).

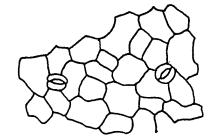
Anisocytic (Cruciferous)

Unequal-celled: three subsidiary cells, one distinctly smaller than the other two, surround the stoma



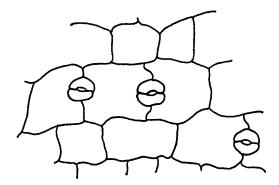
Anomocytic (Ranunculaceous)

Irregular-celled: no subsidiary
cells present



Diacytic (Caryophyllaceous)

Cross-celled: two subsidiary cells enclose the stoma, their common wall at right angles to the long axis of the stoma



Paracytic (Rubiaceous)

Parallel-celled: one or more subsidiary cells occur on either side of the stoma, parallel with its long axis

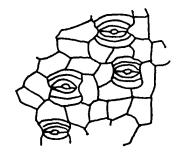
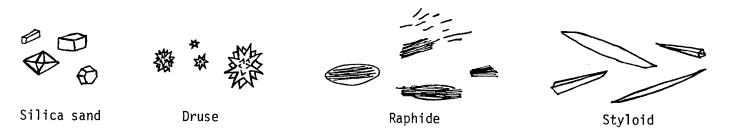


Figure 4. Stomatal arrangement in the leaf epidermis of dicotyledons included in this report (from Fahn 1974).

CRYSTAL FORMS



SILICA BODY SHAPES

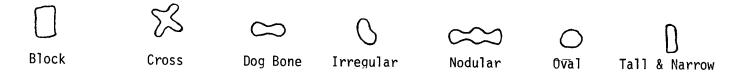


Figure 5. Characteristic forms of crystals and silica bodies observed in selected species included in this report.

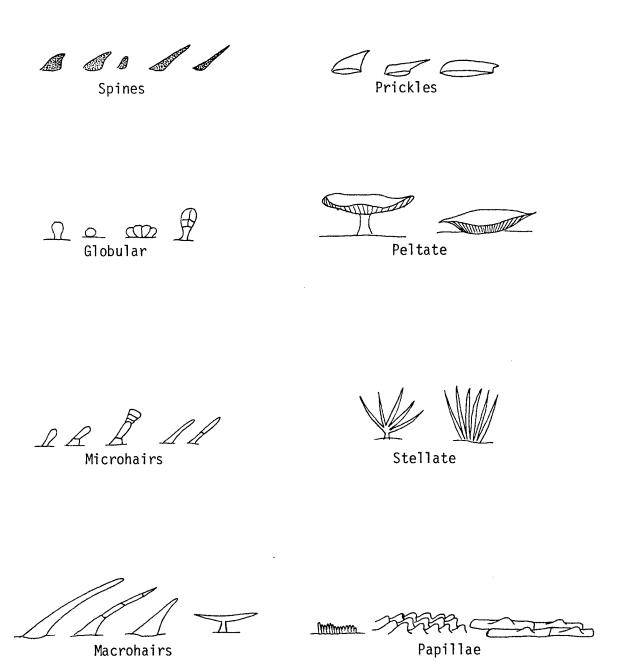
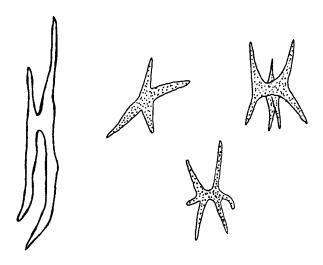


Figure 6. Characteristic forms of trichomes observed in selected species included in this report.



Idioblasts



Tannin Bodies

Figure 7. Characteristic shapes of tannin bodies and idioblasts observed in selected species included in this report.

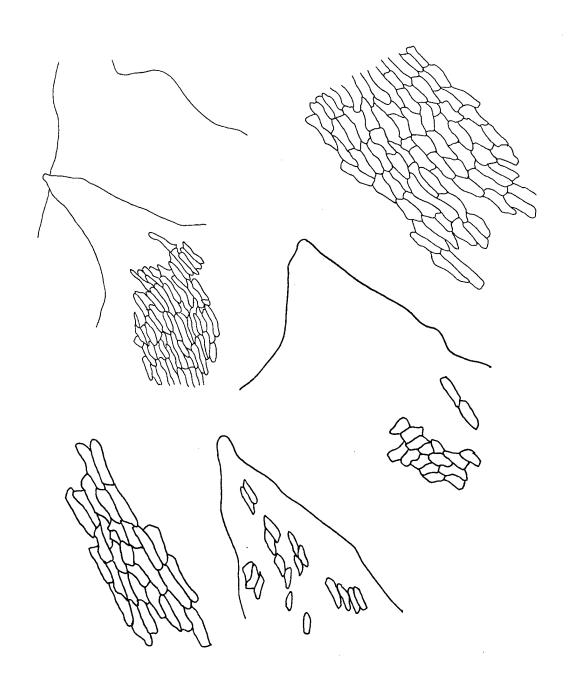


Figure 8. Acanthophora spicifera (Red alga), Family Rhodophyceae. Fragment terete with lobed, irregular cells. Cell length (0.03-0.12 mm) at least 3X width (0.005-0.030 mm). Segments may glow faintly under polarized light. Tips of branches with several short "spines" arranged in a spiral.

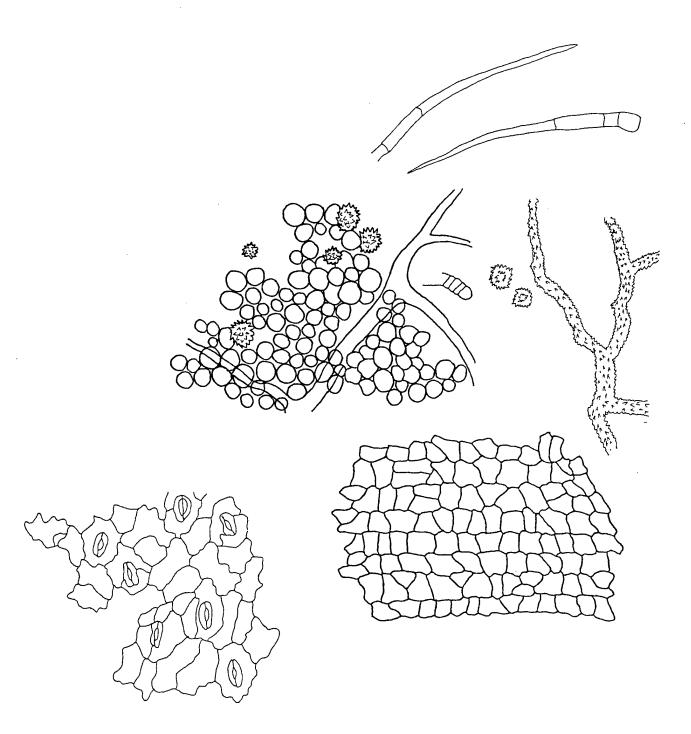


Figure 9. Alternanthera philoxeroides (Alligator weed), Family Amaranthaceae (dicot). Large, solitary druse crystals obvious, common between veins. Veins sometimes completely coated with minute unidentified crystals. Round, globular mesophyll cells obvious beneath very faint, angular epidermal cells 0.02-0.07 mm long, 0.02-0.06 mm wide. Long, multicellular trichomes present in small indentations of leaf margins, though only the basal cell often is present. Short, multicellular trichomes on leaf surface may be present on young leaves. Stomata 0.25 mm long, 0.015 mm wide.

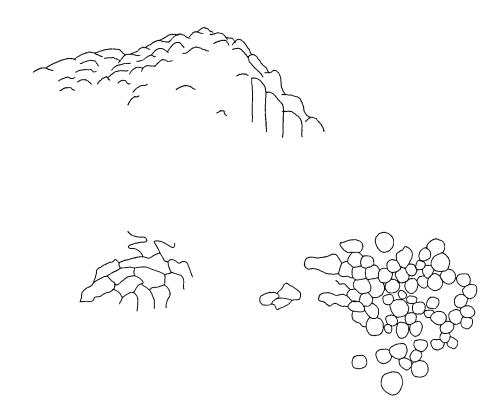


Figure 10. Azolla caroliniana (Waterfern), Family Salviniaceae (fern). Epidermal cells round, 0.02-0.05 mm diameter and papillate, covering small flat "leaflets" (fronds), 0.20-0.50 mm wide and 0.60-0.80 mm long. Vascular tissue sparse, branching into leaflets. Colonies of minute algae (Anabaena) often present inside leaflets.

Some Some

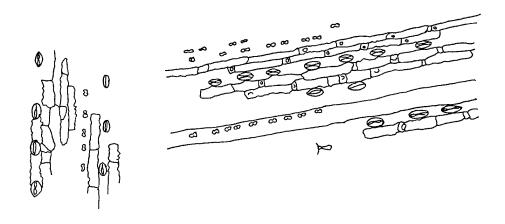


Figure 11. Brachiaria mutica (Grass), Family Gramineae (monocot). Fragment with parallel veins, sinuous cell walls, and silica bodies. Papillae in large, rounded formations, especially obvious along veins, and resembling corn on a cob. Veins one to two cells wide (0.01-0.02 mm wide), narrower than intercostal region, glowing under polarized light. Prickles on upper surface veins numerous. Two to four (usually two) rows of stomata between veins. Stomata 0.020-0.035 mm long, 0.01-0.02 mm wide. Crystal sand and microhairs present. Spines variable in length, present on leaf margins.

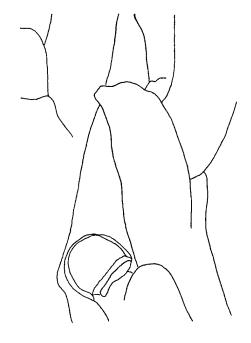


Figure 12. Bugala neritina (Bryozoan), Class Bryozoa (invertebrate). Fragment with branching filaments. Filaments terete, <0.05 mm diameter, without epidermal cells and with numerous attached capsules in regular series; clear to brown-colored, glowing under polarized light. (Drawing at 53X magnification).

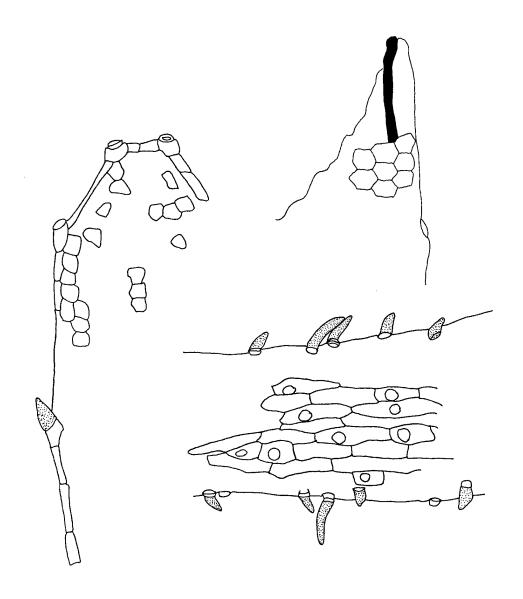


Figure 13. Cabomba caroliniana (Fanwort), Family Nymphaeaceae (dicot). Short, unicellular, deciduous conical spines present on leaflet surface and edge, often evident only by their disk-shaped bases. Epidermal cells angular to lobed, 0.04-0.20 mm long, 0.02-0.04 mm wide. Leaf edge cells often longer than other epidermal cells. Leaflets 0.50-0.90 mm wide. Leaf tips with two spines. No crystals present. Tannin cells observed.

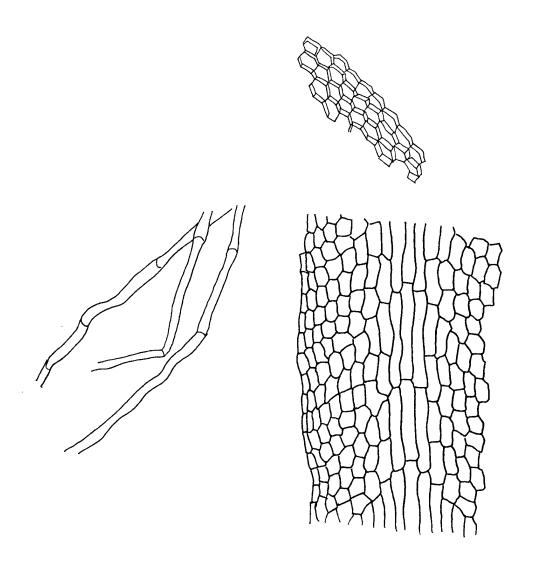


Figure 14. Caloglossa leprieurii (Red alga), Family Rhodophyceae. Cells on edge of blade much smaller than other cells. Midrib obvious on short, narrow (0.5-2.0 mm wide) blade. Cells hexagonal, 0.03-0.20 mm long and 0.01-0.05 mm wide. Blade one cell thick with numerous filaments (rhizoids) extending from constrictions on blades.

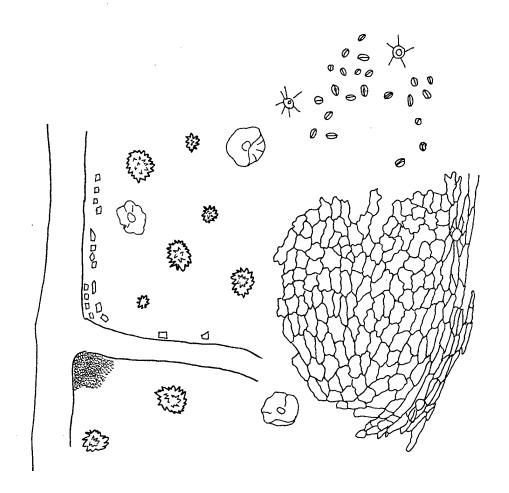


Figure 15. Carya glabra (Pignut hickory), Family Juglandaceae (dicot). Lobed epidermal cells 0.015-0.080 mm long, 0.01-0.04 mm wide, covered with papillae. Venation obvious, reticulate. Numerous large druse crystals between veins. Crystal sand and small druse crystals over veins. Large peltate trichomes abundant. Stomata 0.015-0.020 mm long, 0.010-0.015 mm wide.



Figure 16. Caulerpa prolifera (Green alga), Family Chlorophyceae. Fragment flat with distinct "idioblasts": slender, coiled and worm-like; resembling "Velcro."

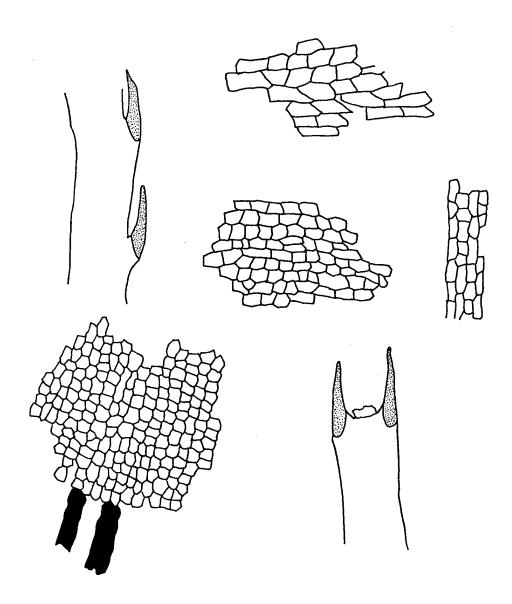


Figure 17. Ceratophyllum demersum (Coontail), Family Ceratophyllaceae (dicot). Epidermal cells mostly angular, 0.02-0.05 mm long, 0.02-0.03 mm wide. Needle-like spines on one margin of leaf only, not abundant. Leaf tip distinct, with two opposing spines. Small, unidentified crystals observed throughout leaf tissue. Tannin cells observed.

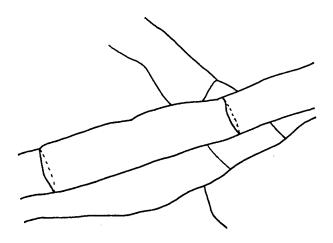


Figure 18. Chaetomorpha brachygona (Green alga), Family Chlorophyceae. Terete, filamentous fragment without epidermal cells. Filaments unbranched, with obvious segments 0.24-0.50 mm long, 0.03-0.14 mm wide. Note: C. brachygona is not distinguishable from Spirogyra sp. by characters listed here.

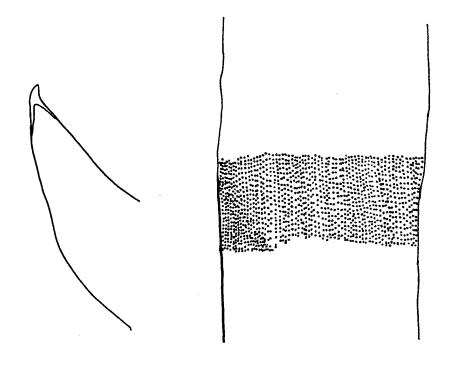


Figure 19. Chara zeylanica (Green alga), Family Charophyceae. Fragment terete, without epidermal cells. Surface pitted, often giving appearance of longitudinal striations. Branching stems and large spines often present at nodes. Segment width 0.30-0.60 mm.

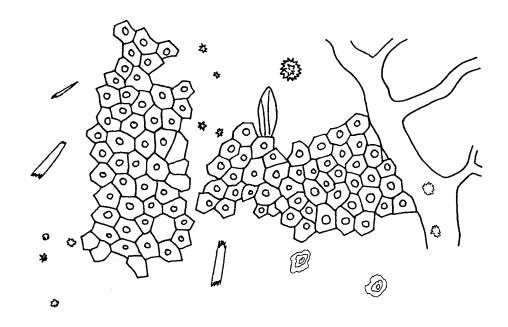


Figure 20. Colocasia esculenta (Wild taro, Elephant ear), Family Araceae (monocot). Raphide and druse crystals numerous and obvious. Venation obvious, reticulate. Epidermal cells angular, 0.010-0.065 mm long, often with a single papilla centered on each cell.

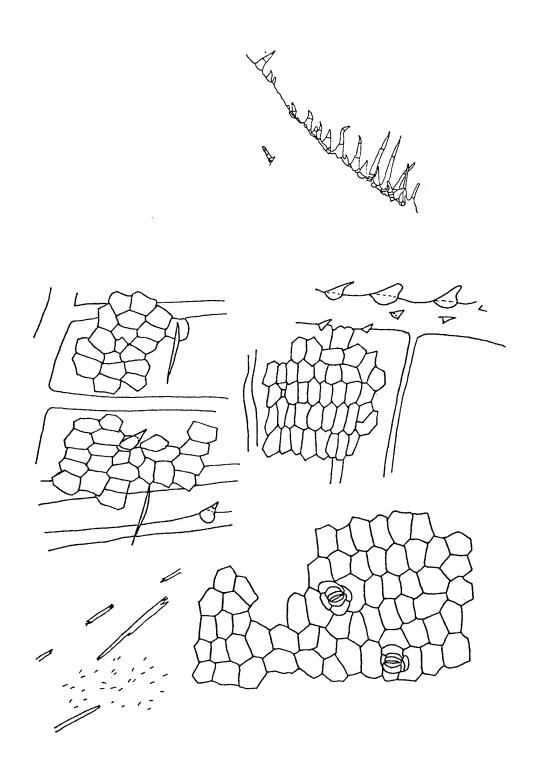


Figure 21. Commelina diffusa (Common dayflower), Family Commelinaceae (monocot). Raphides abundant and very slender, aligned in rows parallel to main veins. Minute crystal sand abundant throughout leaf tissue. Cross veins numerous and parallel to each other. Long veins glow faintly under polarized light, cross veins do not glow. Long and short hook-like prickles and small club-shaped trichomes common on leaf surface and margins. Epidermal cells angular, often hexagonal, 0.015-0.090 mm long, 0.01-0.06 mm wide. Stomata 0.035-0.040 mm long, 0.02-0.04 mm wide.

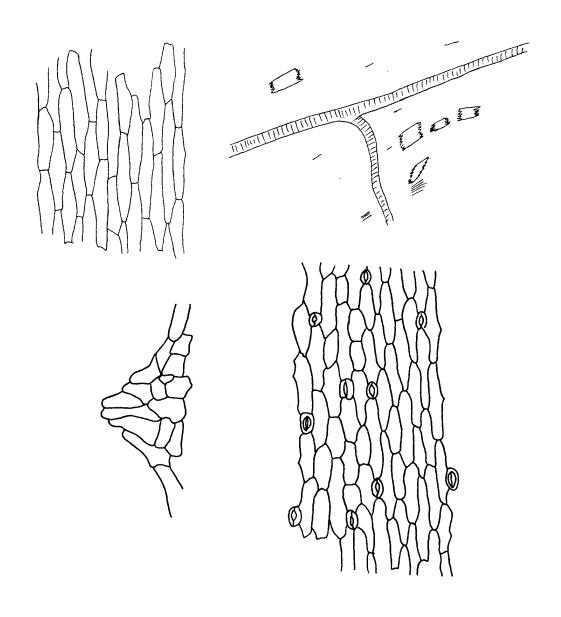


Figure 22. Crinum americanum (Swamp lily), Family Amaryllidaceae (monocot). Raphides solitary, common. Epidermal cells rectangular, lobed to angular, in rows, 0.04-0.24 mm long, 0.01-0.04 mm wide. Stomata numerous, 0.02-0.04 mm long, 0.015-0.030 mm wide. Unidentified trichomes on leaf surface and edge. Leaf fragment may have a gently pleated appearance.

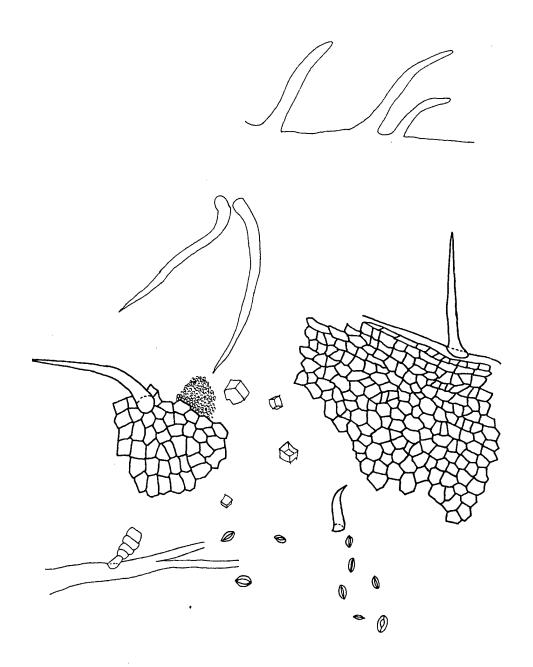


Figure 23. Diospyros virginiana (Common persimmon), Family Ebenaceae (dicot). Veins robust, reticulate. Large crystal sand common, seen as cuboidal blocks. Long, unicellular macrohairs abundant, multicellular club-hairs common. Stomata 0.02-0.03 mm long, 0.010-0.015 mm wide. Angular epidermal cells 0.01-0.05 mm long, 0.01-0.03 mm wide, covered with papillae.

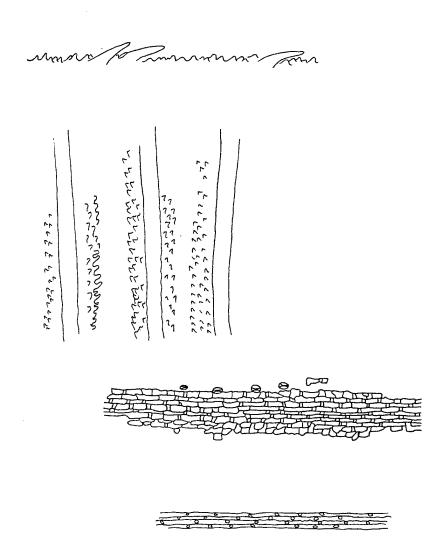


Figure 24. Distichlis spicata (Salt grass), Family Gramineae (monocot). Fragment with large parallel veins, sinuous cell walls, and silica bodies. Veins deep, broad, 0.02-0.07 mm wide, narrower than intercostal region, glowing under polarized light. Two rows of stomata between veins. Stomata 0.015-0.020 mm long, 0.01 mm wide. Large prickles over and between veins. Silica bodies abundant on veins, not in rows but scattered throughout. Papillae small to medium in height, conical to wave-like, more than one per epidermal cell. Papillae obscure all other epidermal features.

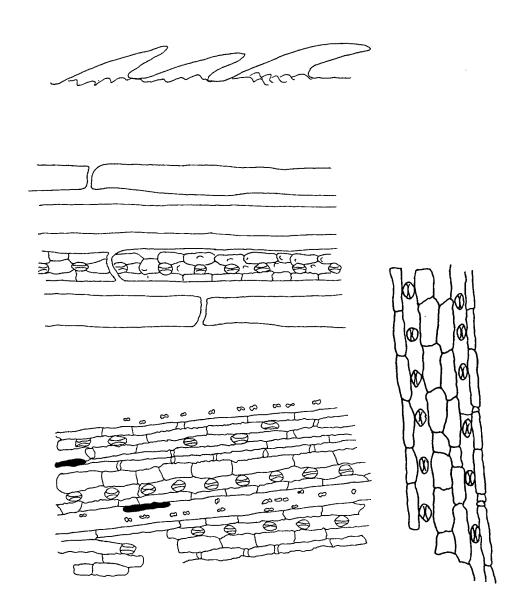


Figure 25. Echinochloa muricata (Watergrass), Family Gramineae (monocot). Fragment with large parallel veins, sinuous cell walls, and silica bodies. Veins 0.01-0.08 mm wide, usually narrower than intercostal region, glowing under polarized light. Veins with one to three rows of silica bodies, dogbone or nodular-shaped over veins, with middle row dogbone-shaped and outer rows tall and narrow, or block to cross-shaped between veins. Prickles present over and between veins. One to three (usually two) rows of stomata between veins, 0.025-0.030 mm long, 0.02 mm wide. One papilla per long cell, wider than tall and appearing as a shallow, wave-like bump. Microhairs rare.

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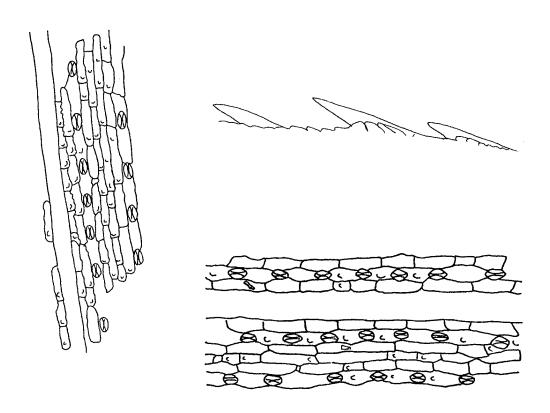


Figure 26. Echinochloa paludigena (Watergrass), Family Gramineae (monocot). Fragment with large parallel veins, sinuous cell walls, and silica bodies. Veins 0.01-0.08 mm wide, usually narrower than intercostal region, glowing under polarized light. Veins with one to three rows of silica bodies, dogbone or nodular-shaped over veins, dogbone-shaped between veins. Prickles present over and between veins. Two rows of stomata between veins, 0.025-0.040 mm long, 0.01-0.02 mm wide. One papilla per long cell, wider than tall and appearing as a shallow, wave-like bump. Microhairs rare.

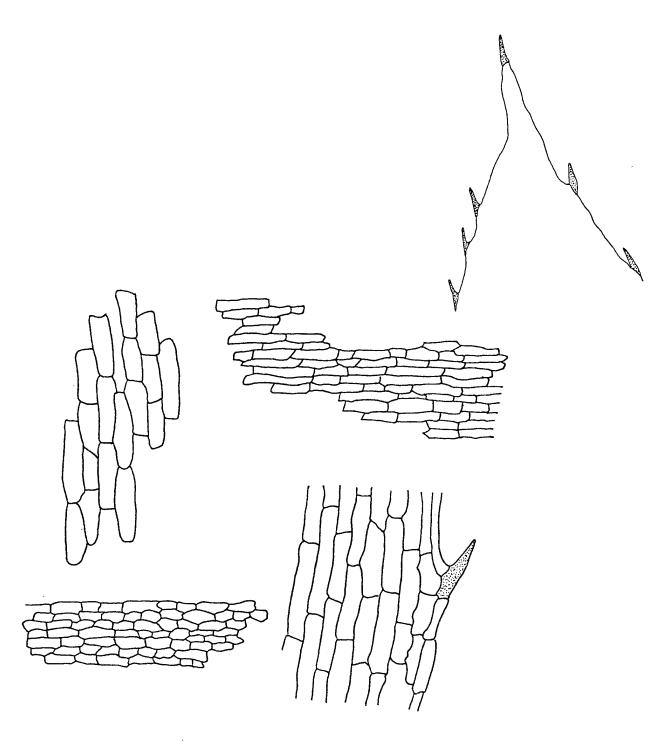


Figure 27. Egeria densa (Brazilian elodea), Family Hydrocharitaceae (monocot). Leaf margins with short, tooth-like spines. Leaf tip with one spine. Midrib vein glows brightly under polarized light. Epidermal cells on the adaxial (upper) surface of the leaf considerably larger than epidermal cells on the abaxial (lower) surface of the leaf. Epidermal cells mostly angular, 0.04-0.17 mm long, 0.01-0.04 mm wide. Leaf edge cells often longer than other epidermal cells. (Leaf tip with spines at 53X magnification).

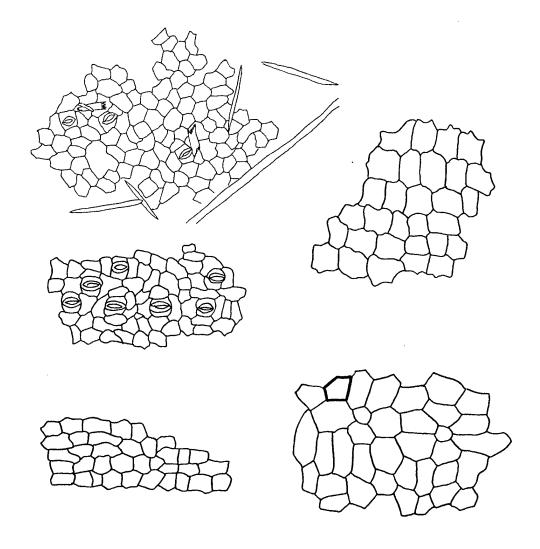
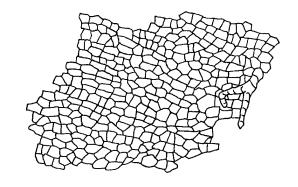


Figure 28. Eichhornia crassipes (Water hyacinth), Family Pontederiaceae (monocot). Main veins parallel, glowing under polarized light; numerous cross veins. Large styloids and smaller raphides equally abundant. Tannin bodies globular, abundant. Epidermal cells angular to lobed, 0.02-0.09 mm long, 0.01-0.06 mm wide. Stomata 0.040-0.045 mm long, 0.020-0.025 mm wide. Note: our limited microscopic observations on Pontederia suggest Eichhornia crassipes and Pontederia cordata lancifolia have nearly identical microhistological features, and are not distinguishable by characters listed here.



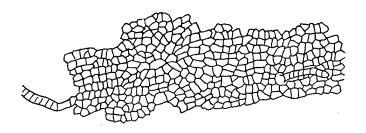


Figure 29. Enteromorpha compressa/Enteromorpha intestinalis (Green alga), Family Chlorophyceae. Green alga with small (0.008-0.050 mm diameter), angular cells. Numerous multicellular filaments extend from terete blade.

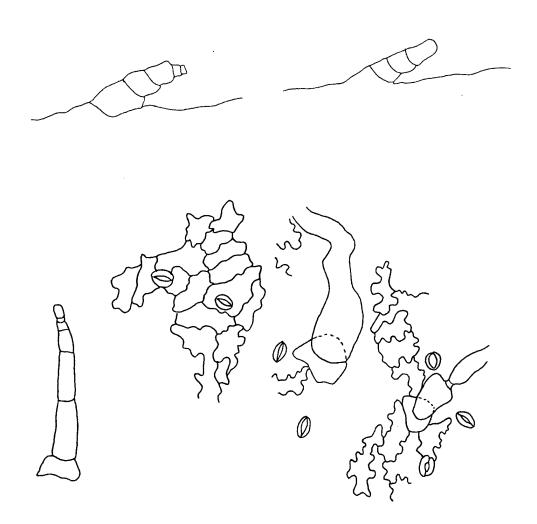


Figure 30. *Erechtites hieracifolia* (Fireweed), Family Compositae (dicot). Venation reticulate with free ends in mesophyll. Epidermal cells lobed to sinuous anticlinal, 0.05-0.12 mm long, 0.015-0.070 mm wide. Stomata present, 0.035-0.045 mm long, 0.02-0.03 mm wide. Multicellular trichomes present. Leaf edge trichomes shorter than leaf surface trichomes.

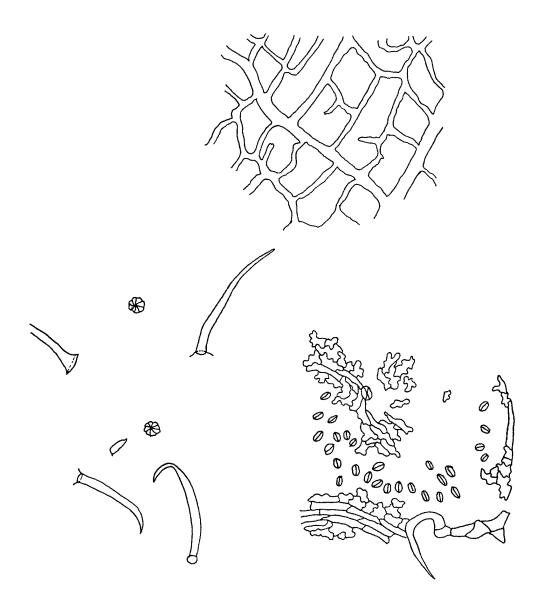


Figure 31. Fraxinus caroliniana (Carolina ash), Family Oleaceae (dicot). Vascular tissue abundant, reticulate. Veins glow brightly under polarized light. Small crystal sand obvious, irregularly abundant along or above veins. Short to long macrohairs common over epidermis. Multicellular peltate hairs small and numerous. Epidermal cells angular to sinuous anticlinal, 0.015-0.090 mm long, 0.01-0.04 mm wide. Stomata 0.015-0.030 mm long, 0.010-0.015 mm wide.

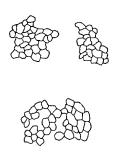


Figure 32. *Gracilaria cervicornis* (Red alga), Family Rhodophyceae. Flat to terete, irregularly or pinnately branching red alga. Fragment with small (0.008-0.020 mm diameter), round or oblong cells.

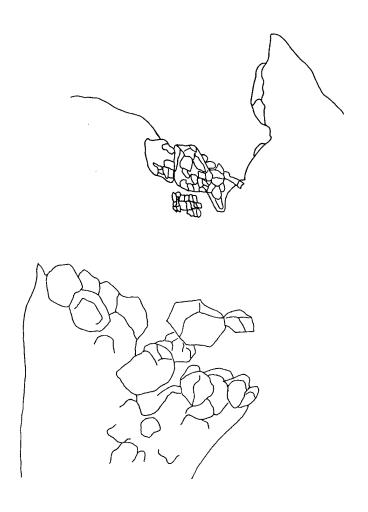


Figure 33. Gracilaria verrucosa (Red alga), Rhodophyceae. Terete, branching alga, purple to red (occasionally gray to green) in color. Branches 0.5-2.0 mm wide. Fragment with two to three layers of small (0.008-0.020 mm diameter) round or oblong cells in cortex. Large round to hexagon-shaped transparent cells frequently visible on surface.

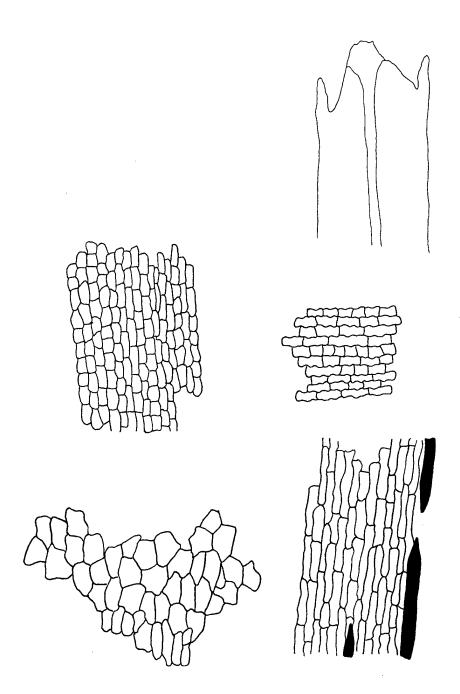


Figure 34. Halodule sp. (Shoal grass), Family Zannichelliaceae (monocot). Leaf blade flat, narrow (≤ 3.0 mm wide), and unbranched. Vascular tissue obvious. Leaf edge veins usually glow brightly under polarized light, more obvious than midrib. Epidermal cells rectangular, 0.02-0.16 mm long, 0.01-0.02 mm wide, often with a distinct, wavy outline, and arranged in rows like bricks. Leaf edge cells same length as other leaf epidermal cells. Leaf tip bi- or tridentate, distinct. Tannin cells common, often pointed at ends and restricted to sides and tip of leaf. Mesophyll not obvious. Leaf material commonly turns reddish-brown after boiling in Hertwig's solution. (Leaf tip at 53X magnification).

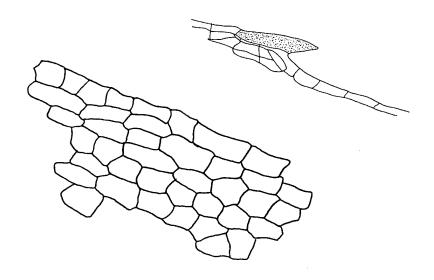


Figure 35. Halophila engelmannii (Halophila), Family Hydrocharitaceae (monocot). Midrib vein and two parallel veins obvious, cross veins less obvious. Leaf margins with tooth-like spines. Leaf tip broad. Epidermal cells on both surfaces of leaf angular to lobed and approximately the same size (0.04-0.11 mm long and 0.03-0.06 mm wide) but leaf edge cells often longer. Vein-glow under polarized light variable. Crystal sand present.



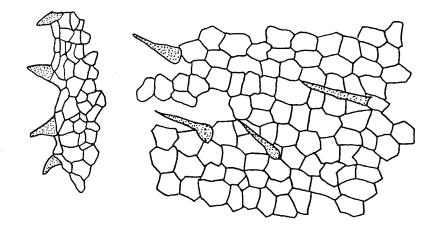


Figure 36. Halophila sp. (Halophila), Family Hydrocharitaceae (monocot). Midrib vein and two parallel lateral veins obvious, cross veins less obvious. Small tooth-like spines common on leaf margins; leaf surface with sparse, short (<0.15 mm long) needle-like spines. All spines are unicellular and small. Epidermal cells angular to lobed, 0.04-0.11 mm long, 0.01-0.04 mm wide. Leaf edge cells similar in size to other epidermal cells. Crystal sand present.

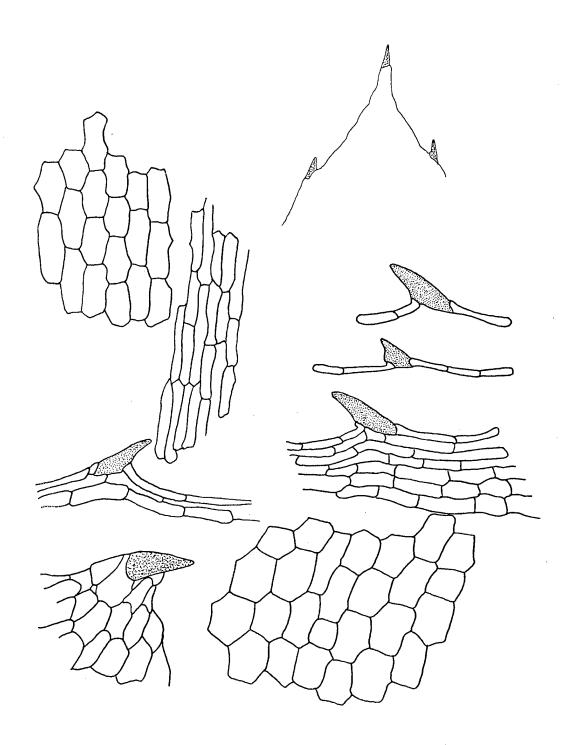


Figure 37. Hydrilla verticillata (Florida elodea), Family Hydrocharitaceae (monocot). Tooth-like spines present on leaf margins. Midrib with infrequent spines. Leaf tip with one spine. Leaf edge and midrib veins glow faintly under polarized light. Epidermal cells on the adaxial (upper) surface of the leaf considerably larger than abaxial (lower) epidermal cells. Epidermal cells angular, 0.04-0.20 mm long, 0.01-0.06 mm wide. Tannin cells often present. Leaf edge cells often longer than other epidermal cells. (Leaf tip with spines at 53X magnification).

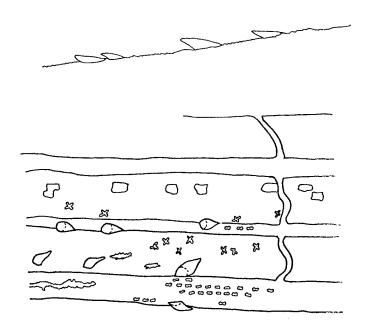


Figure 38. Hydrochloa caroliniensis (Southern watergrass), Family Gramineae (monocot). Fragment with parallel veins, sinuous cell walls, and silica bodies. Veins 0.02-0.09 mm wide, narrower than intercostal region, glowing under polarized light and with one to four rows of dogbone-shaped silica bodies. Intercostal region with numerous cross-shaped silica bodies, distinctive. Cross veins numerous, obvious. Prickles 0.03-0.08 mm long, abundant over and between veins. Two rows of stomata between veins, often difficult to discern. Stomata 0.020-0.025 mm long, 0.01 mm wide. Epidermal cells short and cuboidal on upper surface, long, thin and sinuous on opposite side. Sinuous cells papillate, with abundant small "dots" and larger "bumps". Macro/microhairs rarely seen. Minute crystal sand present.

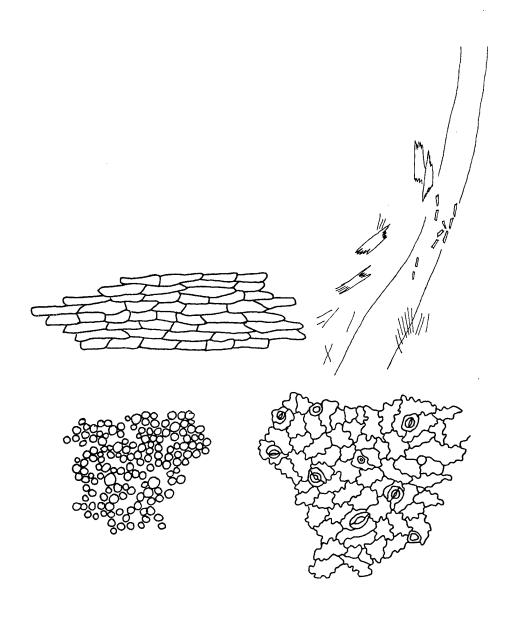


Figure 39. Hydrocotyle sp. (Water pennywort), Family Umbelliferae (dicot). Epidermal cells usually round to oblong, not papillate, and small (0.01-0.02 mm diameter), or occasionally large, sinuous anticlinal, not papillate, and longer than wide (0.03-0.08 mm long, 0.02-0.03 mm wide), covering flat fragment. Vascular tissue abundant, reticulate. Stomata and trichome bases apparent. Stomata 0.020-0.025 mm long, 0.015 mm wide. Styloids, raphides, and crystal sand rare.

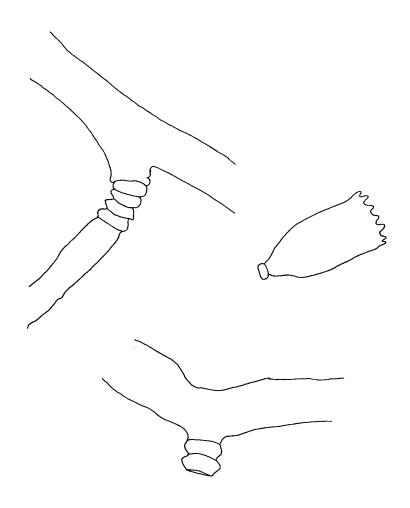


Figure 40. Unidentified hydroid (hydroid), Class Hydrozoa (invertebrate). Fragment of unicellular, branched filaments, with accordion-shaped joints. Filaments brown and terete, without epidermal cells, and <0.5 mm diameter.

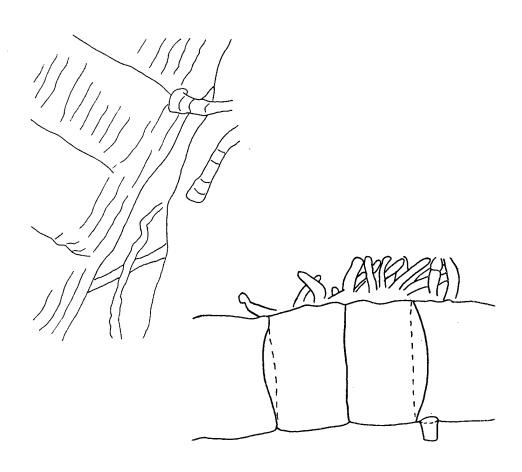


Figure 41. *Hypnea cervicornis* (Red alga), Family Rhodophyceae. Branching, terete, segmented filaments, with numerous minute multicellular "roothairs" extending from surface. Segments wider than long, 0.20-0.40 mm wide, 0.10-0.20 mm long. No epidermal cells.

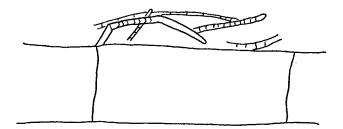


Figure 42. Jania adherens (Red alga), Family Rhodophyceae. Branching, terete, segmented filaments, with numerous minute multicellular "roothairs" extending from surface. Segments longer than wide, 0.30-0.50 mm long, 0.08-0.15 mm wide. No epidermal cells.

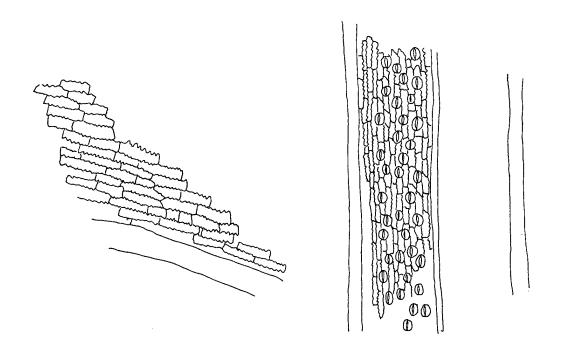


Figure 43. Juncus sp. (Rush), Family Juncaceae (monocot). Fragment with parallel veins and sinuous cell walls. No silica/tannin cells or silica bodies present. Stomata numerous, 0.02-0.03 mm long, 0.015-0.025 mm wide. Veins 0.02-0.10 mm wide, narrower than intercostal region, glowing under polarized light and with six or more rows of stomata between them. Spines, trichomes, and papillae absent.

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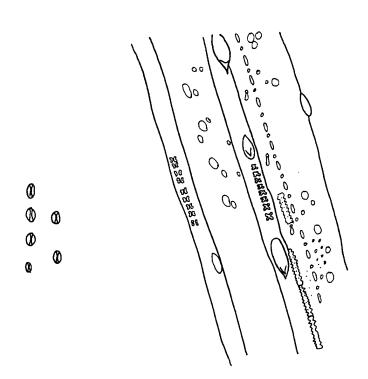


Figure 44. Leersia hexandra (Cut grass), Family Gramineae (monocot). Fragment with large parallel veins, sinuous cell walls, and silica bodies. Veins 0.03-0.06 mm wide, narrower than intercostal region, glowing under polarized light. Main veins separated by two to four rows of stomata. Stomata 0.01-0.04 mm long, 0.005-0.020 mm wide. Dogbone-shaped silica bodies present at right angles to long axes of veins and appearing as two rows of oval bodies between prickles. Silica bodies over veins cross-shaped from edge-view, oblong to dogbone-shaped from above. Prickles present between and above veins. Large prickles over veins abundant. Minute papillae associated with much larger, dome-shaped papillae. Leaf margins bordered with large hook-shaped spines and numerous papillae. Microhairs very small.

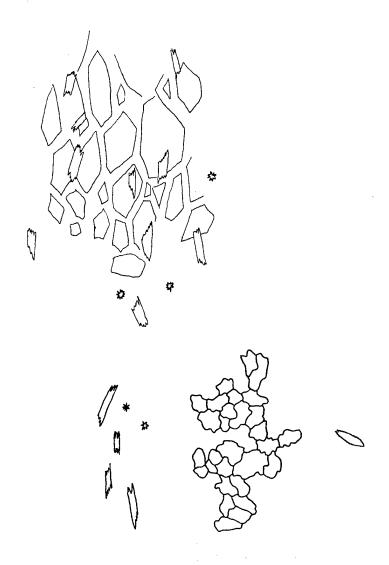


Figure 45. Lemna sp. (Duckweed), Family Lemnaceae (monocot). Epidermal cells lobed to weakly sinuous anticlinal, 0.02-0.05 mm long, 0.02-0.03 mm wide. "Leaflets" (fronds) minute, less than 4.0 mm long, with reticulate venation. Raphides and druse crystals common. Leaf edge cells often longer than other epidermal cells.

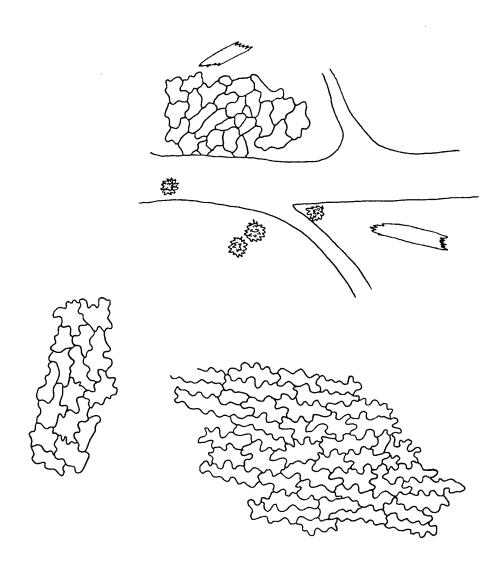


Figure 46. Ludwigia repens (Ludwigia), Family Onagraceae (dicot). Vascular tissue abundant, reticulate, midrib prominent. Epidermal cells irregularly lobed to weakly sinuous anticlinal, 0.02-0.16 mm long, 0.02-0.05 mm wide. Raphides and druse crystals common; crystal sand and styloids also present. Leaf edge cells often longer than other epidermal cells.

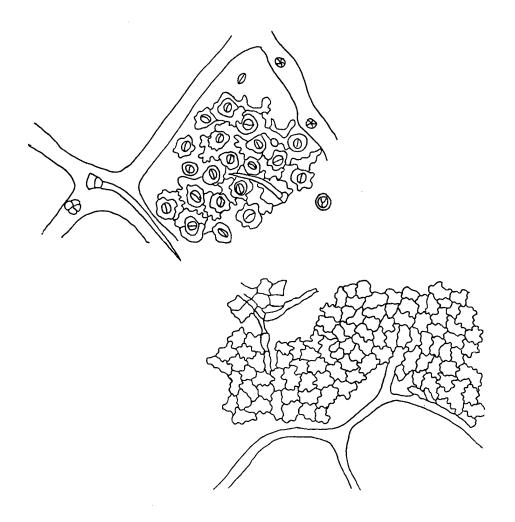


Figure 47. Magnolia grandiflora (Southern magnolia), Family Magnoliaceae (dicot). Vascular tissue abundant, reticulate. Veins glow faintly under polarized light. Trichomes generally small, globular, multicellular, abundant. Few long multicellular macrohairs also present. Epidermal cells strongly sinuous anticlinal (jigsawed), 0.02-0.08 mm long, 0.01-0.07 mm wide. Stomata abundant, 0.02 mm long, 0.01-0.02 mm wide. Unidentified spherical bodies (possibly oil droplets) locally abundant.

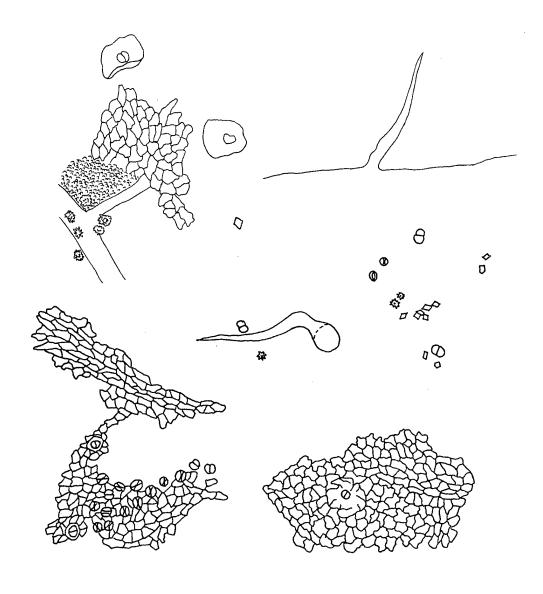


Figure 48. Myrica cerifera (Wax myrtle), Family Myricaeceae (dicot). Angular to weakly sinuous anticlinal epidermal cells, 0.01-0.04 mm long, 0.01-0.02 mm wide, covered with papillae. Stomata small, 0.020-0.025 mm diameter. Venation reticulate but usually difficult to discern. Crystal sand and small druse crystals above veins, infrequently between veins. Large peltate and macrohair trichomes common on leaf surface. Macrohairs present on leaf edge, abundant along midvein.

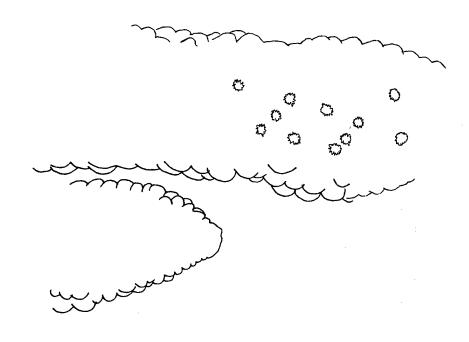


Figure 49. Myriophyllum aquaticum (Parrot's feather), Family Haloragaceae (dicot). Epidermal cells round, 0.02-0.06 mm diameter, papillate, covering small terete leaflets 0.15-0.35 mm wide. Vascular tissue difficult to discern. Solitary druse crystals are common and aligned along cortex inside leaflets.

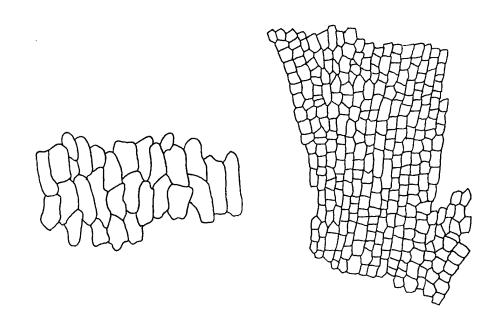


Figure 50. Myriophyllum spicatum (Eurasian watermilfoil), Family Haloragaceae (dicot). Fragment terete to flat, highly branched, with leaflets (pinnae) 0.15-0.30 mm wide, attached to a stalk (rachis) 0.25-0.70 mm wide. Epidermal cells angular, generally block-shaped, 0.015-0.080 mm long and 0.01-0.02 mm wide, and arranged in rows. Druse crystals uncommon, restricted to cortex of main stem.

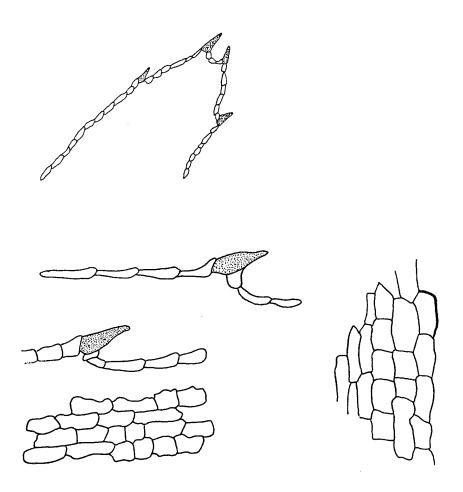


Figure 51. Najas guadalupensis (Southern naiad), Family Najadaceae (monocot). Epidermal cells on both surfaces of leaf approximately the same size, 0.04-0.12 mm long, 0.02-0.05 mm wide, angular, arranged in rows. Leaf edge cells often longer than other epidermal cells. Leaf edge veins glow brightly under polarized light. Leaf tip with one or two spines; leaf margins with tooth-like spines. (Leaf tip with spines at 53X magnification).

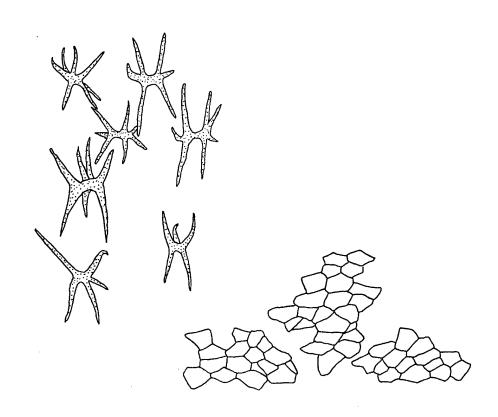


Figure 52. Nuphar luteum (Spatterdock), Family Nymphaeaceae (dicot). Fragment flat with irregularly stellate idioblasts, concentrated along major vascular bundles. Idioblasts sometimes covered with minute crystals and glow brightly under polarized light. Vein-glow variable under polarized light. Epidermal cells round, approximately 0.02-0.05 mm diameter.

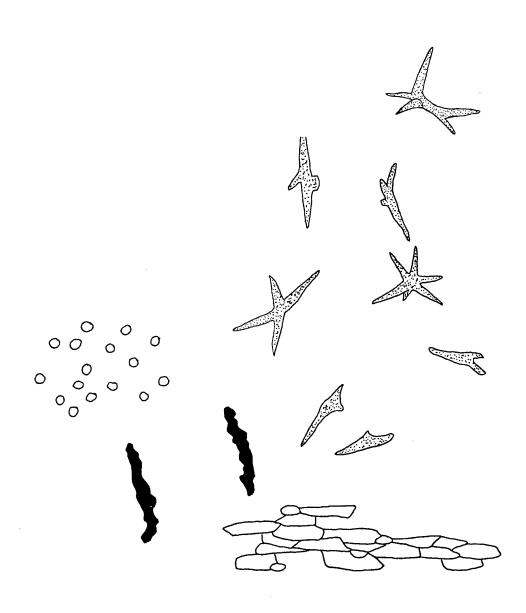


Figure 53. Nymphaea mexicana (Yellow waterlily), Family Nymphaeaceae (dicot). Fragment flat with numerous irregularly stellate idioblasts obvious throughout leaf tissue. Idioblasts sometimes covered with minute crystals and glow brightly under polarized light. Vein-glow variable under polarized light. Epidermal cells round, approximately 0.02-0.05 mm diameter. Leaf edge cells often longer than other epidermal cells.

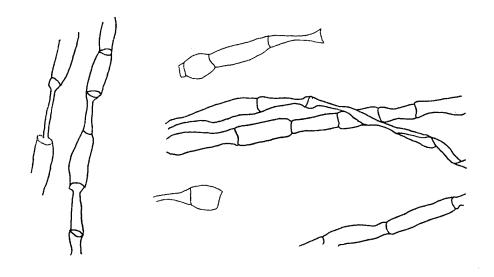
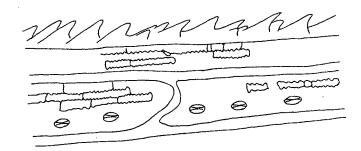
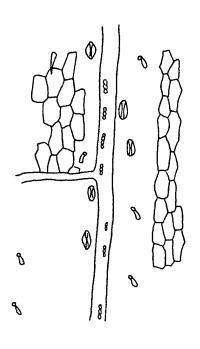


Figure 54. *Oedogonium* sp. (Green alga), Family Chlorophyceae. Filamentous, terete fragment without epidermal cells. Filaments unbranched and with obvious segments, 0.10-0.15 mm long and 0.005-0.050 mm wide.





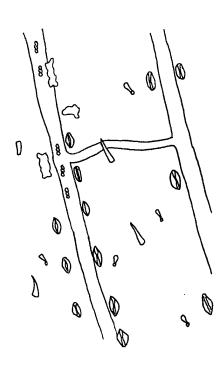
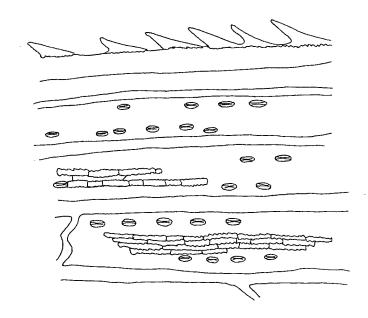


Figure 55. Oplismenus setarius (Grass), Family Gramineae (monocot). Fragment with parallel veins, sinuous cell walls, and silica bodies. Veins 0.01-0.03 mm wide and narrower than intercostal region, glowing under polarized light and separated by two rows of stomata. Stomata 0.02-0.04 mm long, 0.01-0.02 mm wide. Macrohairs abundant between veins. Microhairs club and hair-shaped. Occasional cross-shaped silica bodies between veins. Silica bodies over veins nodular and dogbone-shaped, mostly in one row (middle vein of leaf has three rows). Prickles infrequent, 0.020-0.025 mm long between veins and 0.04-0.08 mm long above veins. Spines abundant and overlapping on leaf margins. Papillae absent.

Figure 56. Oscillatoria tenuis (Brown alga), Family Cyanophyceae. Fragment terete, filamentous, very fine and hair-like, 0.005-0.010 mm diameter, without epidermal cells. Many small segments are very difficult to see on the unbranched filaments. (Drawing at 266X magnification).



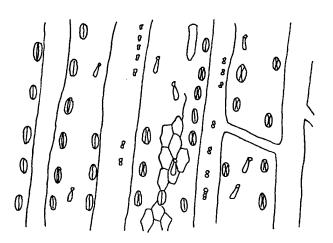


Figure 57. Panicum commutatum (Grass), Family Gramineae (monocot). Fragment with parallel veins, sinuous cell walls, and silica bodies. Veins narrower than intercostal region, 0.02-0.09 mm wide, glowing under polarized light and with one row of cross or dogbone-shaped silica bodies (except for middle vein, which has three rows) over veins. Veins separated by two to three rows of stomata. Stomata 0.02-0.03 mm long, 0.010-0.025 mm wide. One row of unidentified objects (possibly prickles) between veins; prickles on veins common. Microhairs club-shaped. Spines numerous on leaf margins. Papillae absent.

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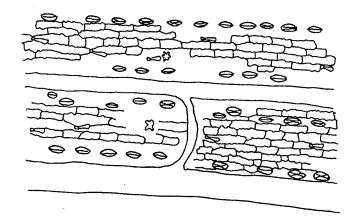


Figure 58. Panicum dichotomum (Grass), Family Gramineae (monocot). Fragment with parallel veins, sinuous cell walls, and silica bodies. Veins 0.02-0.06 mm wide, narrower than intercostal region, glowing under polarized light and separated by two rows of stomata. Stomata 0.030-0.045 mm long, 0.01-0.02 mm wide. Microhairs often with long, distal cell, usually same size as basal cell. One row of unidentified objects (possibly prickles) between veins; prickles on veins common. Silica cell pairs few. Epidermal cells very sinuous. Spines numerous on leaf margins. Papillae absent.

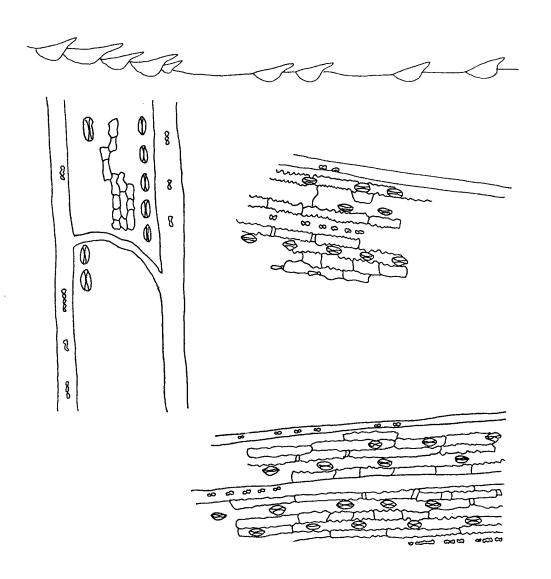


Figure 59. Panicum hemitomon (Maidencane), Family Gramineae (monocot). Fragment with parallel veins, sinuous cell walls, and silica bodies. Veins 0.01-0.04 mm wide, much narrower than intercostal region, glowing under polarized light and separated by one to two rows of stomata. Stomata 0.02-0.04 mm long, 0.01-0.02 mm wide. Silica bodies on veins dogbone to nodular-shaped. One row of silica bodies between veins, irregular, mostly tall and narrow, sometimes crenate. Microhairs club and hair-shaped. Spines numerous on leaf margins. Prickles on veins common. Papillae absent.



Figure 60. Paspalum distichum (Seashore paspalum), Family Gramineae (monocot). Fragment with numerous large parallel veins, sinuous cell walls, and silica bodies. Leaf epidermis with huge, distinct papillae, more than one per epidermal cell. Papillae abundant and especially obvious on leaf margins where they form a continuous series of deeply lobed to very broad wave-like formations. Culm material with abundant short cells in pairs. Silica bodies mostly oval between veins, sporadic; dogbone, nodular, or irregular over veins. Veins very high in profile and glow under polarized light; 0.02-0.18 mm wide (wider than intercostal region) on papillate side and 0.02-0.04 mm wide (narrower than intercostal region) on opposite side. Micro-prickles present between veins. Club and hair-shaped microhairs present but often obscured by papillae. Two rows of stomata between veins. Stomata 0.02-0.03 mm long, 0.01-0.02 mm wide.





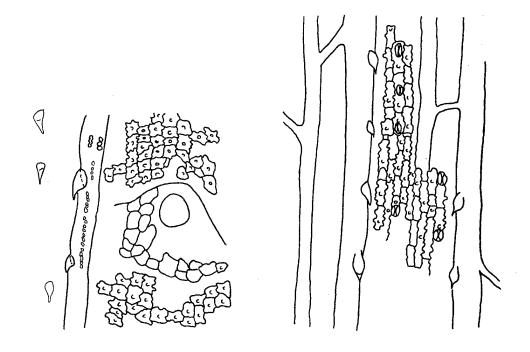


Figure 61. Paspalum repens (Water paspalum), Family Gramineae (monocot). Fragment with numerous large parallel veins, sinuous cell walls, and silica bodies. Epidermal cells block-shaped, with one nipple-shaped papilla in center of each cell; papillae taller than wide. Culm material with abundant cross-shaped silica bodies, much like Hydrochloa leaf epidermis. Veins 0.02-0.08 mm wide, narrower than intercostal region, and glow under polarized light. Leaf material with abundant prickles on veins (0.04-0.10 mm long) and between veins (0.02-0.06 mm long). Macrohairs very large; microhairs present. Two rows of stomata between main veins. Stomata 0.02-0.04 mm long, 0.01-0.04 mm wide. Nodular silica bodies on veins in one to three rows. Spines and papillae evident on leaf margins.

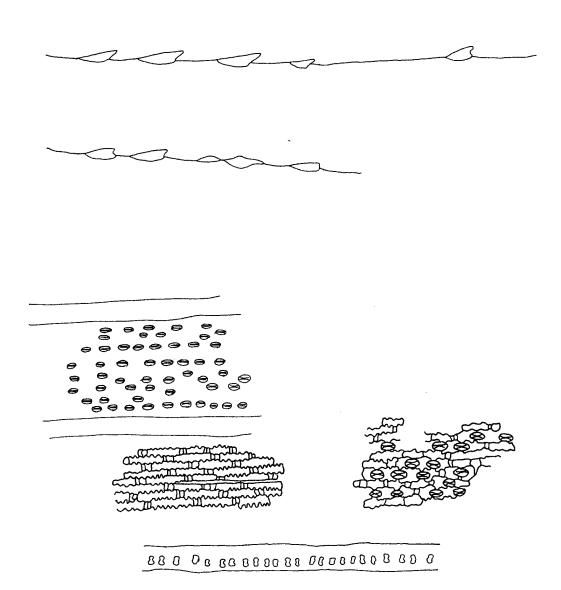


Figure 62. Phragmites australis (Giant reed), Family Gramineae (monocot). Fragment with parallel veins, sinuous cell walls, and silica bodies. Silica bodies block to oblong-shaped, abundant. Veins 0.02-0.08 mm wide, narrower than intercostal region, glowing under polarized light. Stomata numerous between veins, in approximately ten or more rows. Stomata 0.03-0.04 mm long, 0.02 mm wide. Papillae absent. Microhairs present. Leaf surface prickles observed on leaf tip only; 0.01-0.06 mm long in intercostal region, 0.06-0.10 mm long over veins. Spines present on leaf margins.

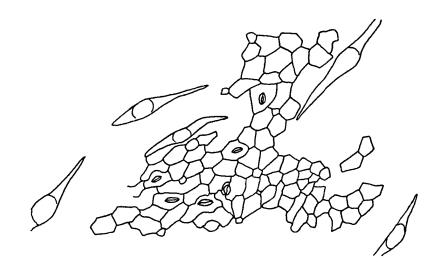
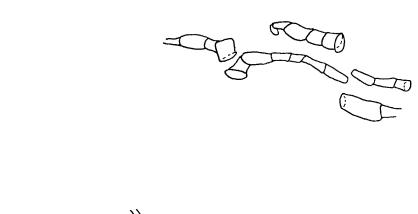


Figure 63. *Phyla nodiflora* (Frog fruit), Family Verbenaceae (dicot). Fragment with numerous, large, T-shaped trichomes that resemble idioblasts. Epidermal cells angular to lobed, 0.01-0.07 mm long, 0.01-0.05 mm wide. Stomata 0.015-0.025 mm long, 0.005-0.015 mm wide.



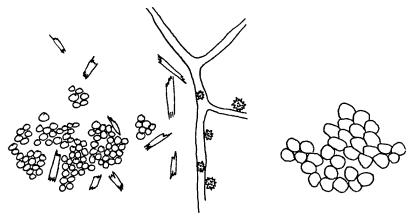


Figure 64. *Pistia stratiotes* (Water lettuce), Family Araceae (monocot). Epidermal cells round, bubble-like, 0.010-0.015 mm diameter. Numerous multicellular trichomes (possibly epiphytic algae) on one side of leaf. No stomata. Venation reticulate but often difficult to discern. Druse crystals frequently on cross veins. Both druse and raphide crystals abundant in mesophyll.

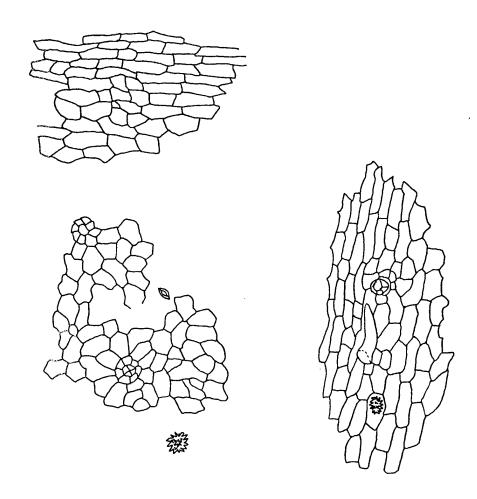


Figure 65. *Polygonum punctatum* (Smartweed), Family Polygonaceae (dicot). Solitary druse crystals present. Multicellular globular and unicellular hair trichomes present. Narrow, leaf edge spines present. Epidermal cells angular to lobed, 0.01-0.10 mm long, 0.01-0.05 mm wide. Stomata 0.020-0.025 mm long, 0.015 mm wide.

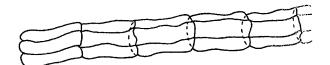


Figure 66. *Polysiphonia subtilissima* (Red alga), Family Rhodophyceae. Terete, filamentous fragment without epidermal cells. Filaments branched and segmented, with each segment composed of four long cylindrical cells. Segments joined end-to-end in a cylindrical formation. Segments 0.10-0.20 mm long, 0.04-0.07 mm wide.

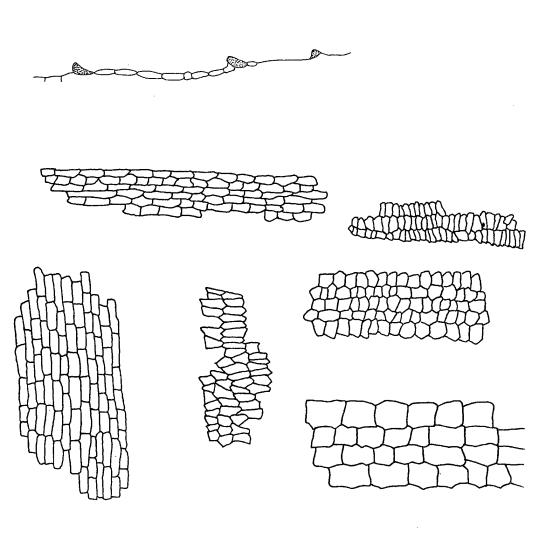


Figure 67. Potamogeton illinoensis (Illinois pondweed), Family Potamogetonaceae (monocot). Leaf blade >3.0 mm wide; veins numerous. Parallel and cross veins glow faintly under polarized light. Leaf edge veins glow brightly as a thin line. Midrib vein very broad relative to lateral veins. Leaf edge spines minute and deciduous, often not present. Stomata and crystals absent. Epidermal cells in linear series and variably block to irregularly rectangle-shaped, 0.01-0.08 mm long, 0.005-0.050 mm wide.

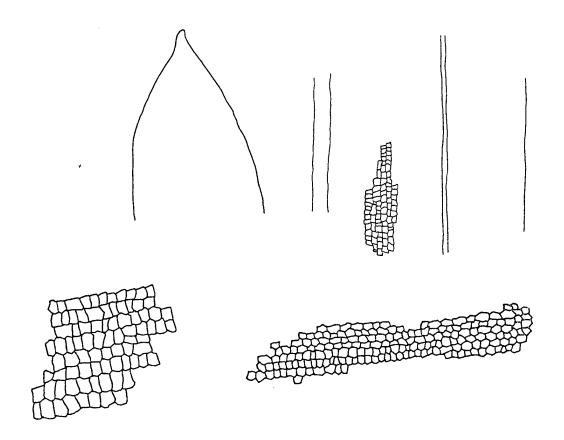


Figure 68. Potamogeton pectinatus/Potamogeton pusillus (Grassleaf pondweed, Sago pondweed), Family Potamogetonaceae (monocot). Leaf blade 0.5-3.0 mm wide; veins obvious, but few. Broad midrib with two lateral veins and leaf edge veins. Leaf edge vein glows brightly as a thin line under polarized light; midrib and lateral veins glow less brightly. Cross veins present, connecting center vein to only one side vein at a time. Epidermal cells 0.005-0.040 mm long, 0.005-0.030 mm wide, rectangular to block-shaped. Leaf edge cells often longer than other leaf epidermal cells. Stomata and crystals absent. Mesophyll easily seen below epidermis. Leaf tip acuminate. (Leaf tip at 53X magnification).

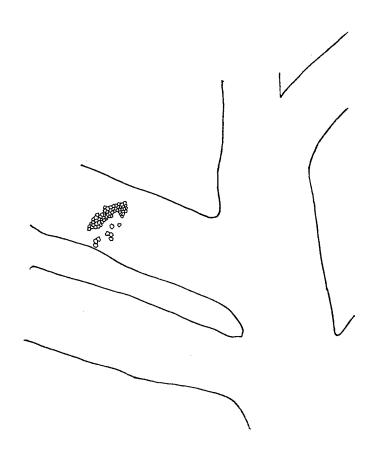


Figure 69. Pterocladia americana (Red alga), Family Rhodophyceae. Branching red alga with unsegmented filaments and minute (0.0025-0.0100 mm diameter), angular cells that are difficult to discern. Blade width 0.10-0.60 mm.

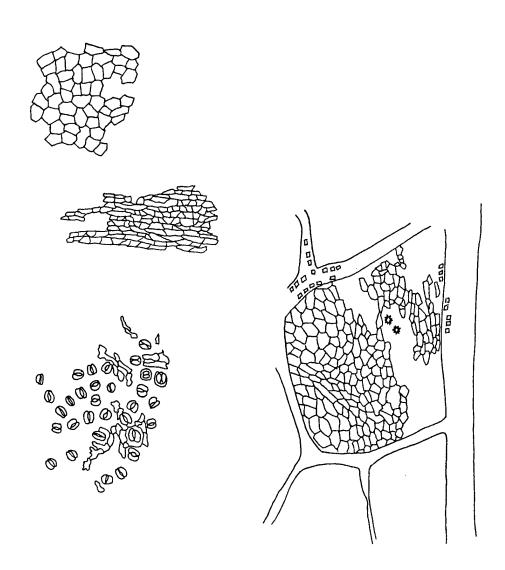


Figure 70. Quercus nigra (Water oak), Family Fagaceae (dicot). Vascular tissue abundant, reticulate, with veins glowing under polarized light. Crystal sand occurring on veins, druse crystals in tissue between veins. Angular to lobed epidermal cells 0.01-0.05 mm long, 0.01-0.03 mm wide. Stomata 0.020-0.025 mm long, 0.01-0.02 mm wide, numerous.

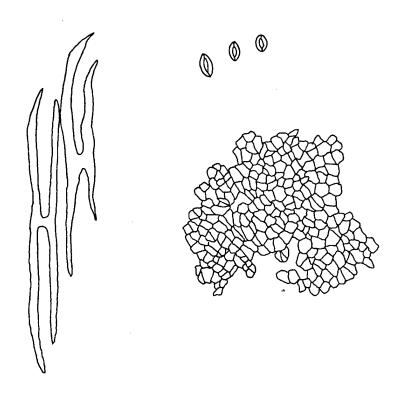


Figure 71. *Rhizophora mangle* (Red mangrove), Family Rhizophoraceae (dicot). Large distinct H-shaped idioblasts common; druse crystals abundant in tissue. Both idioblasts and crystals glow brightly under polarized light. Angular to lobed epidermal cells, 0.010-0.035 mm long, 0.005-0.020 mm wide. Stomata 0.03-0.04 mm long, 0.020-0.025 mm wide. Tannin cells observed.

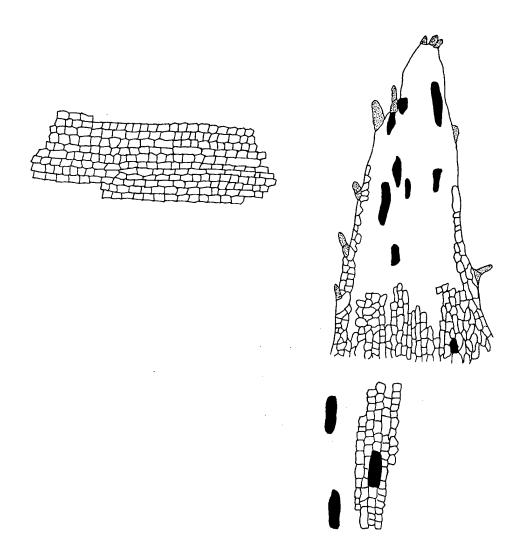


Figure 72. Ruppia maritima (Widgeon grass), Family Ruppiaceae (monocot). Veins not obvious. Leaf blade flat, unbranched, narrow, <3.0 mm wide. Epidermal cells mostly angular, small, arranged in rows; 0.01-0.03 mm long, 0.01-0.02 mm wide. Leaf edge cells same length or shorter than other epidermal cells. Secretory cells (possibly empty tannin sacs) common. Tannin bodies common, oval to rectangular, evenly distributed on leaf surface. Mesophyll not apparent. Leaf edge spines short and irregularly spaced, restricted mostly to tip of leaf. Leaf tip acute, with small teeth.

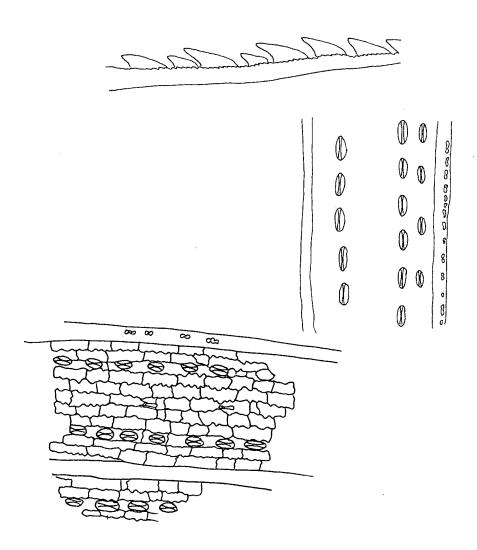


Figure 73. Sacciolepis striata (Grass), Family Gramineae (monocot). Fragment with numerous parallel veins, sinuous cell walls, and silica bodies. Silica bodies large, nodular to dogbone-shaped on veins, not apparent in intercostal region. Veins 0.02-0.06 mm wide, narrower than intercostal region, glowing under polarized light. Veins separated by two to four rows of stomata. Stomata 0.035-0.050 mm long, 0.015-0.030 mm wide. Stomata subsidiary cells mostly low dome-shaped, but also triangular. Paired short cells uncommon. Leaf edge with small bumps (papillae?) and large spines. Crystal sand common. Microhairs club-shaped. Prickles on veins uncommon.

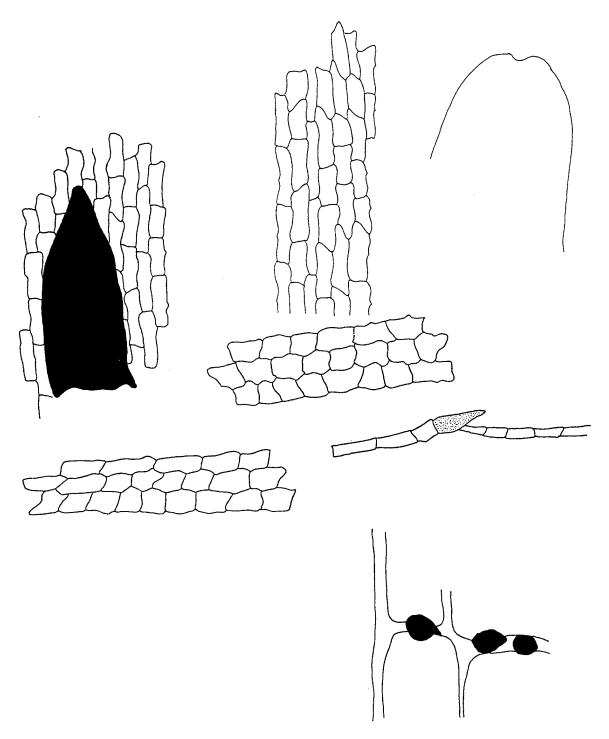


Figure 74. Sagittaria kurziana/Sagittaria stagnorum (Strap leaf, Water arrowhead), Family Alismataceae (monocot). Leaf blade >3.0 mm wide. Veins numerous, parallel longitudinal veins usually glowing under polarized light. Epidermal cells in linear series, large, block-shaped or often hexagonal, 0.03-0.14 mm long, 0.02-0.05 mm wide. Leaf edge spines rare (S. kurziana) to absent (S. stagnorum). Prickles occasionally present. S. kurziana with globular trichomes. Stomata and crystals absent. Tannin bodies uncommon.

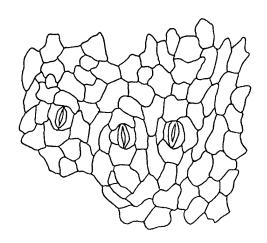


Figure 75. Sagittaria lancifolia (Duck-potato), Family Alismataceae (monocot). Leaf blade >3.0 mm wide. Veins and cross veins numerous, usually glowing under polarized light. Stomata 0.04-0.05 mm long, 0.02-0.03 mm wide. Unidentified crystals common. Epidermal cells lobed to angular, 0.03-0.10 mm long, 0.02-0.05 mm wide, not in linear series.

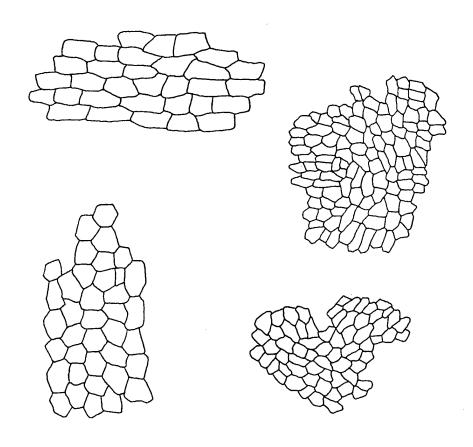


Figure 76. Sagittaria subulata (Arrowroot), Family Alismataceae (monocot). Leaf blade >3.0 mm wide. Veins numerous, parallel longitudinal veins usually glowing under polarized light. Epidermal cells in linear series, large, block-shaped or often hexagonal, 0.030-0.010 mm long, 0.01-0.05 mm wide. Leaf edge spines absent. Crystal sand occasionally present. Tannin bodies uncommon.

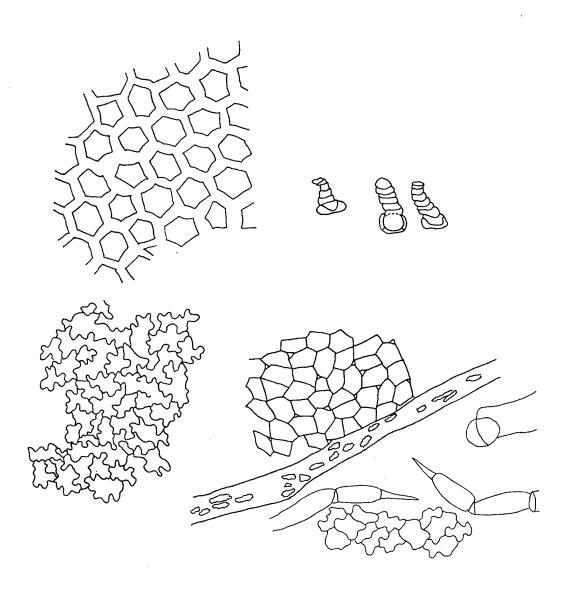


Figure 77. Salvinia rotundifolia (Common salvinia), Family Salviniaceae (fern). Venation reticulate. Epidermal cells distinctly sinuous anticlinal (jigsawed) or very angular, 0.03-0.09 mm long, 0.01-0.05 mm wide. Leaf edge cells often longer than other epidermal cells. Crystal sand often in main veins. Multicellular trichomes (possibly epiphytic algae) short with many cells or long with few cells and a spine-like tip, common on leaf surface. No stomata. "Leaflets" (fronds) small, oval.

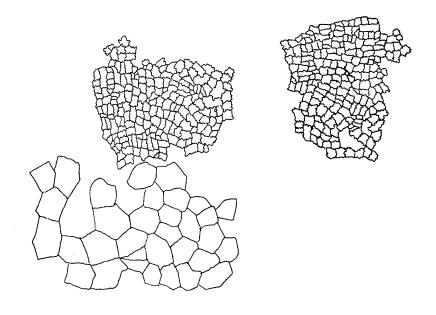


Figure 78. Sargassum filipendula (Brown alga), Family Phaeophyceae. Blade broad, ≥ 3.0 mm wide. Blade edge usually serrate. Epidermal cells irregularly lobed to sinuous anticlinal, small (0.015-0.035 mm long, 0.01-0.02 mm wide), and in a net-like arrangement. Midrib vein obvious. Round bladders at ends of short stalks occasionally present on fragment.

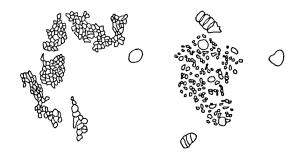


Figure 79. Soliera tenera (Red alga), Family Rhodophyceae. Fragment terete with minute (0.001-0.015 mm diameter) oblong epidermal cells, often with associated unidentified (possibly tannin) bodies, 0.02-0.04 mm diameter.

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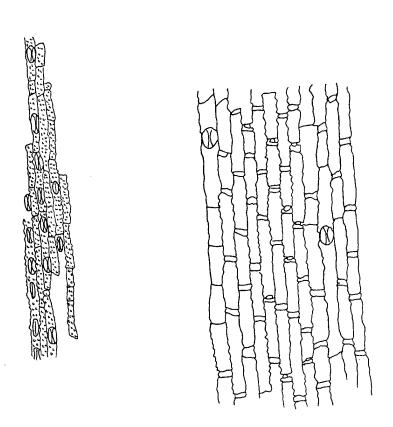


Figure 80. Spartina alterniflora (Saltwater cordgrass), Family Gramineae (monocot). Fragment with large parallel veins, sinuous cell walls, and silica bodies. Veins very fibrous, 0.08-0.40 mm wide, often wider than intercostal region, high in profile, glowing under polarized light, and covered on one side with abundant minute papillae. Silica bodies small, irregular to oval-shaped, not abundant. Silica bodies not seen in or on veins, but silica cells seen easily through vein from opposite side of epidermis. Silica cells (possibly cork cells) between veins tall, narrow and irregularly crenate to rectangular, numerous. Prickles uncommon, difficult to see. Stomata uncommon, intermittently spaced in two rows between veins. Stomata 0.015-0.030 mm long, 0.01-0.02 mm wide. Narrow-based spines common on leaf margins.

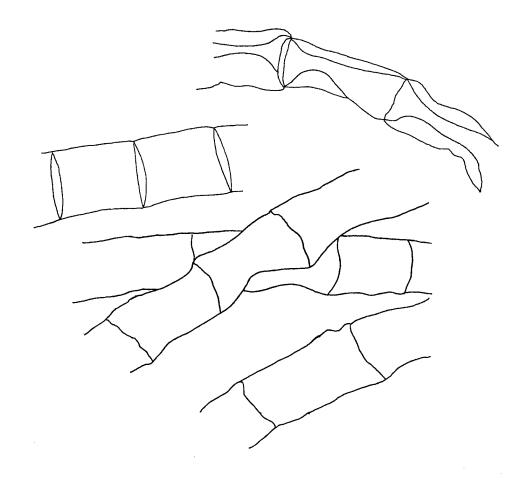


Figure 81. *Spirogyra* sp. (Green alga), Chlorophyceae. Terete, filamentous fragment without epidermal cells. Filaments unbranched and with obvious segments 0.24-0.50 mm long, 0.03-0.14 mm wide. <u>Note:</u> *Spirogyra* sp. is not distinguishable from *Chaetomorpha brachygona* by characters listed here.

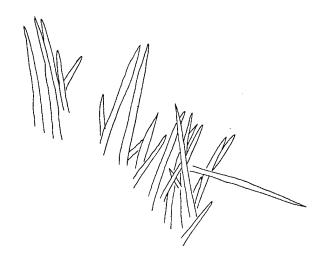


Figure 82. Unidentified sponge (sponge), Class Porifera (invertebrate). Fragment with obvious "idioblasts" (spicules), long and needle-like; non-glowing under polarized light.

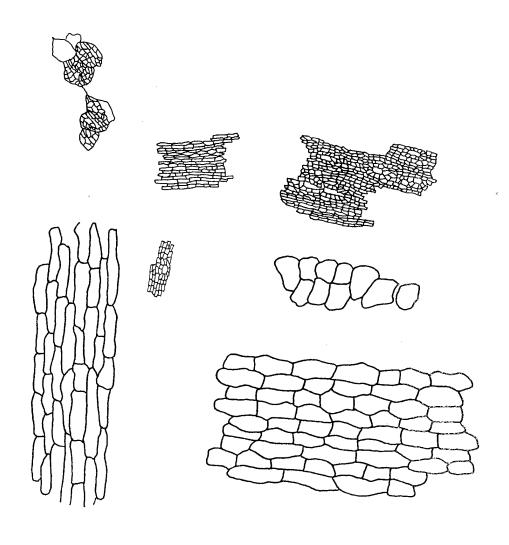


Figure 83. Syringodium filiforme (Manatee grass), Family Zannichelliaceae (monocot). Epidermal cells minute (0.010-0.015 mm long, 0.005-0.010 mm wide), irregularly rectangle to oblong-shaped, and arranged in regular rows. Shadows and extensions of large bulbous cells in the underlying mesophyll give the epidermis a distinct "bumpy" appearance. Numerous secretory cells with narrow surface pores bordered by wide, angular cells. Leaf terete, unbranched.

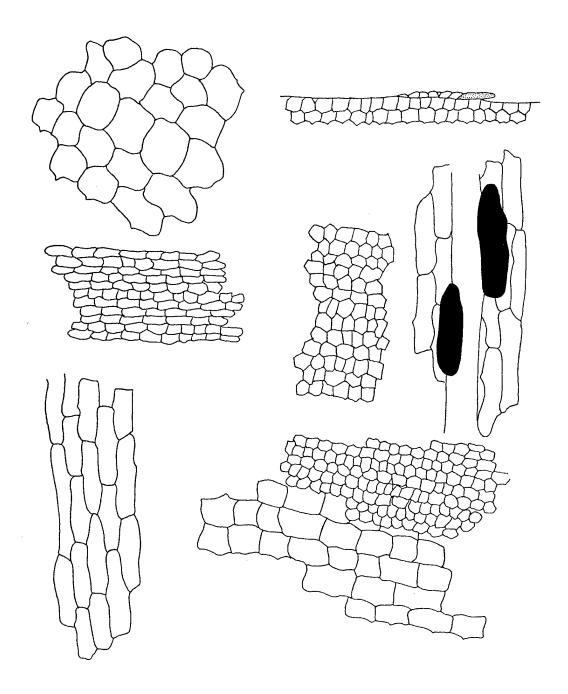


Figure 84. Thalassia testudinum (Turtle grass) Family Hydrocharitaceae (monocot). Leaf blade >3.0 mm wide. Vascular tissue obvious with numerous parallel veins of equal size, very fibrous, glowing under polarized light. Cross veins do not glow. Leaf tip with numerous irregular teeth. Leaf edge spines uncommon. Epidermal cells arranged in linear series, block-shaped, angular to lobed, 0.015-0.050 mm long, 0.01-0.03 mm wide. Tannin cells beneath epidermis, variable in shape. Stomata absent. Crystal sand seen occasionally.

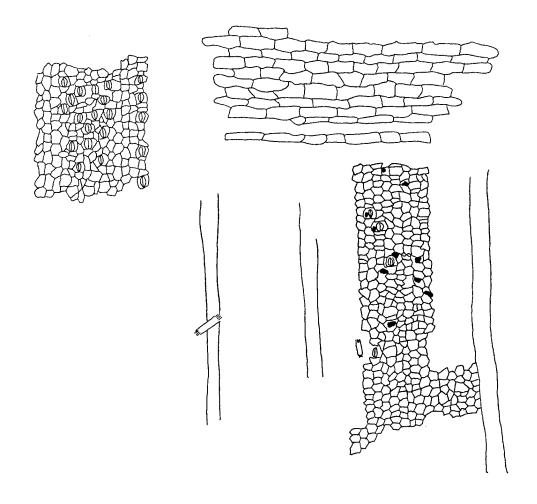


Figure 85. Typha domingensis (Southern cattail), Family Typhaceae (monocot). Fragment with numerous parallel veins that glow (often multicolored) under polarized light. Raphides common. Crystal sand variable in abundance, small. Tannin bodies numerous, small and globular. Epidermal cells angular to lobed, 0.01-0.03 mm long. Five or more rows of stomata between veins. Stomata 0.015-0.030 mm long, 0.010-0.015 mm wide.

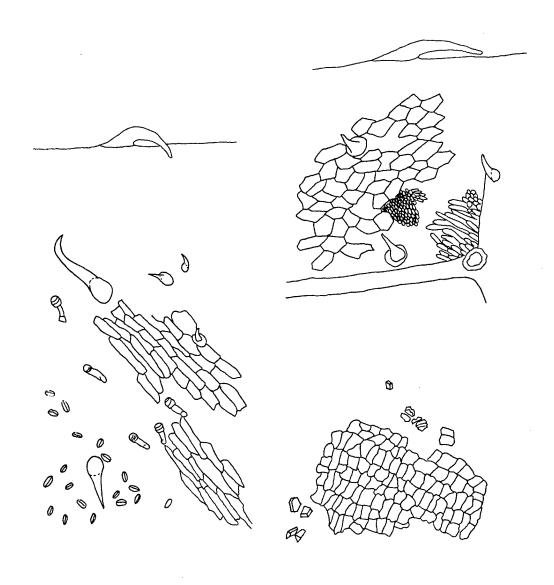


Figure 86. Ulmus americana (American elm), Family Ulmaceae (dicot). Vascular tissue abundant, reticulate. Trichomes consist of short and long prickles, small club-shaped hairs, and large macrohairs. Trichome bases often hexagonal in shape. Prickles and macrohairs most common over veins. Crystal sand small, uncommon. Epidermal cells angular to lobed, 0.02-0.14 mm long, 0.01-0.04 mm wide, covered with papillae. Stomata 0.02 mm long, 0.01 mm wide.

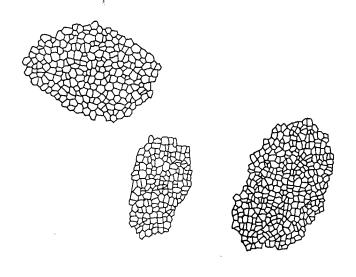


Figure 87. Ulva lactuca (Green alga), Family Chlorophyceae. Blade thin, flat, entire, with small (0.010-0.025 mm long, 0.005-0.025 mm wide), angular cells.

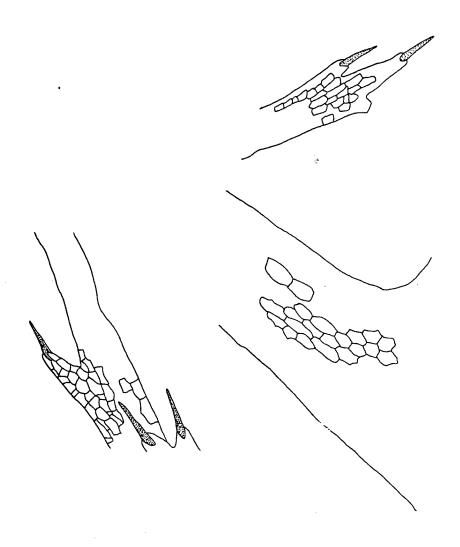


Figure 88. *Utricularia* sp. (Bladderwort), Family Lentibulariaceae (dicot). Leaf margins with short, needle-like spines, alternately opposite; leaf usually tipped with one spine. Round bladders present occasionally along edges of leaves. Epidermal cells angular, 0.015-0.070 mm long, 0.01-0.03 mm wide.

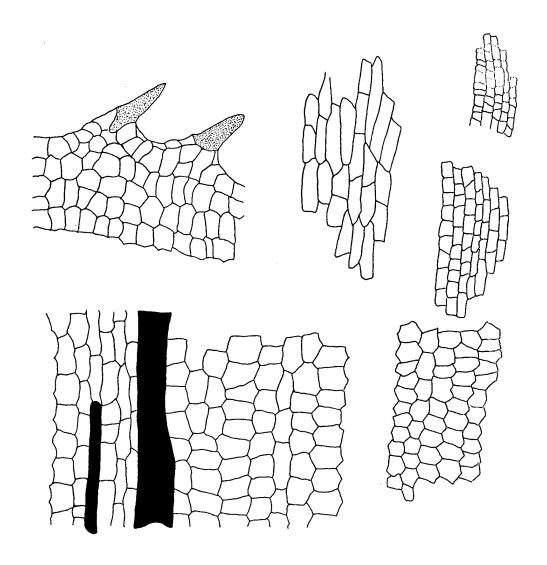


Figure 89. Vallisneria americana (Tapegrass, Eelgrass), Family Hydrocharitaceae (monocot). Leaf blade broad, >3.0 mm wide. Parallel veins numerous, usually glowing under polarized light. Cross veins not obvious. Leaf edge spines robust, common. Epidermal cells block to hexagon-shaped, large, 0.03-0.06 mm long, 0.02-0.05 mm wide, and arranged in rows. Leaf edge cells same length or shorter than other epidermal cells. Tannin bodies globular to filamentous, variable in number. Globular trichomes rare. Stomata and crystals absent.

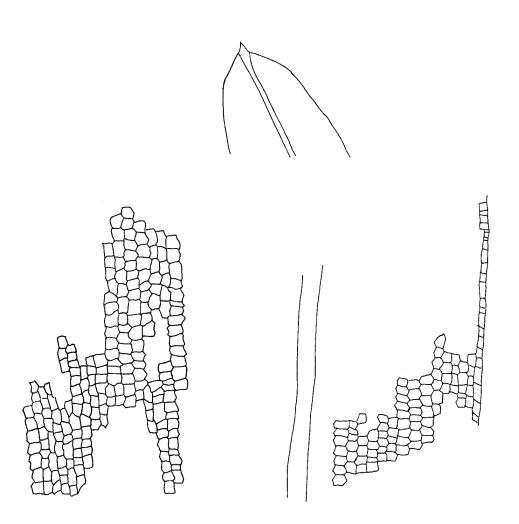


Figure 90. Zannichellia palustris (Horned pondweed), Family Zannichelliaceae (monocot). Leaf blade flat, unbranched, and narrow, ≤3.0 mm wide. Veins few with broad midrib more obvious than leaf edge veins. Cross veins sometimes apparent. Veins do not glow under polarized light. Epidermal cells arranged in rows like bricks, block-shaped, often hexagonal, 0.02-0.03 mm long, 0.01-0.02 mm wide. Leaf edge cells same length as other epidermal cells. Mesophyll apparent. Leaf tip pointed, without spines. (Leaf tip at 53% magnification).

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APPENDIX A. MATERIALS NECESSARY FOR MANATEE INGESTA ANALYSIS

MATERIALS

plant press herbarium paper dissecting needles forceps Hertwig's solution Hoyer's solution household bleach Histoclad (Clay Adams Co.) microscope with 100x magnification and graduated mechanical stage polarizing light filter microscope slides (75 x 25 mm) cover slips (22 x 50 mm, # 1.5) alcohol or gas flame water or 70% alcohol (diluting agent) reference plant collection reference slide collection reference photomicrographs

METHODS OF PLANT PRESERVATION

Plants can be preserved either dry (traditional herbarium method) or wet. Specimens preserved dry offer compact, light storage and easy access. Specimens preserved wet require much space and leak-proof containers but retain their shape and form well. Both methods provide adequate material for reference slides, but preference should be given to fresh material whenever possible.

USE AND DIRECTIONS FOR PREPARATION OF MOUNTING MEDIA

Hoyer's Solution

200 g chloral hydrate crystals

50 ml water

20 ml glycerin

30 g <u>flaked</u> gum arabic (powdered gum arabic does not dissolve well)

Used for mounting plant material on slides; water soluble. Combine and mix over low heat until ingredients dissolve.

Hertwig's Solution

19 ml 1 molar hydrochloric acid

150 ml water

60 ml glycerin

270 g chloral hydrate crystals

Used for clearing plant material before observation. Add acid to water, then combine and mix ingredients over low heat until completely dissolved.

Plant Preservative

3400 ml water

124 ml 36% aqueous formaldehyde 18 ml 20% detergent solution

7 ml saturated copper sulfate solution

Used for preserving and storing plants in liquid.

APPENDIX B. RECOMMENDED METHOD FOR ANALYSIS OF MANATEE INGESTA

Direct observation, utilization studies, ingesta analysis, and fecal analysis are procedures used to estimate the botanical composition of the diet of herbivores. The relevance of these methods to food habits studies of manatees is explained briefly below.

The most direct and simplest method of determining diet composition is by direct observation. Several field studies of the Florida manatee have recorded feeding behavior and food preferences (Hartman 1979; Bengtson 1983; Etheridge et al. 1985), but none have quantified the diet. Direct observation of manatees in the field is practical in Florida only with cold-weather-induced aggregations of relatively tame animals in clear, warm-water refugia or by closely following radiotagged individuals in suitable habitat. However, these situations are restrictive, and unless vegetation occurs in single-species stands identification of ingested material remains problematic.

Utilization studies are traditional approaches to the study of forage consumption in other herbivores. General observations, evaluation of differences before and after grazing, and comparison of grazed and ungrazed plots are some of the methods used in utilization studies. These procedures provide information on locations and quantities of vegetation being used. Unfortunately, these techniques have severe limitations. Unknown rates of plant growth and loss of forage to weathering, human disturbance, or other herbivores all contribute to inaccurate results in evaluating forage consumption. Holechek et al. (1982) reviewed utilization studies and recommended not using them for diet determination.

Fecal analysis is a popular procedure in food habits studies. Some advantages are: (1) non-interference with the animal's habits, (2) relatively unlimited sampling, (3) unrestricted animal movement, (4) allowance for comparison of two or more animals at the same time or longitudinal monitoring of single individuals, and (5) simple collection methods. Unfortunately, disadvantages are formidable, and include: (1) inaccuracy due to discriminate digestion of certain forage plants, (2) disproportional fragmentation of food during digestion, and (3) the rendering of some plant species unidentifiable in the feces (Holechek et al. 1982).

Ingesta analysis is used commonly in food habits studies. This procedure involves analysis of forage collected from the mouth, stomach, intestine, or other portion of the digestive tract. Sacrifice of the animal is usually necessary in this approach. Fistula and trocar techniques have been used for collecting ingesta samples from some species. However, the need for tranquilization and surgery prevents their use on rare and endangered wild animals. Analysis of manatee ingesta is the most practical method in Florida,

however, because of the existence of an active manatee carcass salvage program (Bonde et al. 1983). At present this method of dietary analysis is recommended.

The available methods for botanical analysis of ingesta and fecal samples from an herbivore may be classified into four categories: (1) macroscopic appraisal, (2) manual separation with weight or volume analysis, (3) microhistological analysis, and (4) microscope point analysis (Theurer et al. 1976).

Macroscopic appraisal of ingesta involves qualitative estimates of the botanical composition of the animal's diet (Theurer et al. 1976). Cook et al. (1958) determined that macroscopic appraisal can be used to identify most browse plants. Species are identified by gross morphological characteristics such as leaf shape, size, texture, color, and venation. Grasses and forbs, however, are generally masticated beyond recognition for macroscopic appraisal.

The manual separation technique employs the macroscopic appraisal procedure for separating the specific food items of the ingesta into distinct samples. Each sample is then weighed and analyzed according to need. This technique is useful only when ingesta samples consist of large food fragments easily identified and manipulated by hand. Johnstone and Hudson (1981) used the manual separation technique to analyze ingesta samples taken from the mouths of dugong carcasses in New Guinea. They reported the occurrence and relative abundance of seagrasses, algae, and mangrove in 102 samples of unmasticated to partially masticated ingesta. The dugong's relatively simple diet of seagrasses, algae, and mangroves enabled investigators to analyze mouth ingesta samples by hand. However, the technique used by Johnstone and Hudson (1981) is not suitable for analysis of manatee diet in Florida because the manatee's diet is more complex, usually consisting of freshwater and marine angiosperms, algae, and emergent vascular plants. Many of these plants are difficult to identify by visual appraisal in the field, let alone after mastication. Quantification of manatee ingesta, whether from the mouth, stomach, or intestine, requires examination of microstructure for identification of plant fragments.

Microhistological analysis is a favored method for diet study of herbivores (Holechek and Vavra 1981). This method, as reported by Sparks and Malechek (1968), involves oven-drying and then grinding the ingesta sample through a 1.0-mm screen to reduce all fragments to a uniform size. The milled ingesta is washed over a 200-mesh screen to remove dirt and very small fragments, and insure mixing. The ingesta mixture is then subsampled onto five slides and cleared with Hertwig's and Hoyer's solutions. Vavra and Holechek (1980) reported that soaking the ingesta in sodium hydroxide before clearing increased the number of identifiable particles. Twenty locations are systematically observed at 125X magnification for each slide. Epidermal fragments other than trichomes are recorded as evidence of a plant species for each location. All identifiable particle fragments are recorded in each field. Frequency percentages of each species are converted to particle density per field using the table from Fracker Percent relative density is calculated from particle and Brischle (1944). density to estimate directly the percentage dry weight of each species occurring in the ingesta sample (Sparks and Malechek 1968).

Using ingesta from esophageally fistulated heifers on forest and grassland range, Holechek and Vavra (1981) analyzed the effects of the number of slides examined and frequency of observation on the precision of microhistological analysis. Following ingesta analysis procedures of Sparks and Malechek (1968), they determined that one slide per sample of ingesta would achieve a confidence level within 10% of the sample mean at the 95% probability level for species constituting 30% or more of the sample weight. To achieve the same precision for species composing 20% or more of the sample weight, nine slides would be needed per sample. Minor species of the ingesta sample would require 60 slides to obtain the same level of precision. These authors determined that a minimum of 20 frequency observations per slide, with five slides per sample, would give reasonable estimates of botanical composition for species composing 20% or more of the ingesta sample.

The microscope point technique is another procedure for analyzing ingesta samples quantitatively. Theurer et al. (1976) reported this technique as a modification of the point method first used by Levy and Madden (1933) for identifying species composition of pastures. Galt et al. (1982) describe the standard microscope point technique as illustrated by Harker et al. (1964) and used by Galt et al. (1969). A random sample of ingesta is washed, air-dried, and then placed in a 45×15 cm tray to cover the bottom to about 3 mm in depth. The tray is examined under a microscope at 20% to 80% magnification. At each point, the plant hundred microscope points are taken per sample. fragment appearing immediately under the eyepiece cross hair is identified and recorded. Percent points per sample are based on a total number of identifiable The unidentifiable portion of the sample is assumed to be points recorded. proportional to the identifiable portion. Galt et al. (1982) determined the percent points of a species to be approximately equal to percent volume. Density constants (g/cc) are then developed for each species to convert percent points to percent weight (Galt et al. 1982). Galt et al. (1968) reported that 400 microscope points per sample would achieve a precision within 5% of the mean at the 90% level of probability for species constituting 30% to 60% of the sample Species composing 10% to 30% of the sample weight would be estimated within 20% of the mean at the 90% level of probability. Confidence limits were found to be considerably greater when fewer than 400 points per sample were recorded, while increasing the points recorded beyond 400 resulted in little improvement of precision in relation to the increase of analytical time.

Channels and Morrissey (1981) analyzed dugong stomach contents using a modified microscope point technique. In their procedure, eight slides were prepared from each stomach sample by spreading a random portion of the ingesta evenly over each slide. The slides were first scanned to identify species present in the ingesta. Next, ten sites on each slide were examined systematically at established stops to quantify the species present. Identification of food fragments was done at 40% to 100% magnification. Each field of view had a total of 42 points defined by the ends of 21 lines in a Weibel graticule. The percentage of each species was calculated by counting the number of plant fragments intercepted by the end-points of the lines. They reported on the occurrence and percent composition of food items in 95 dugong

stomachs. Ingesta was identified to the generic level as well as type (leaves and stems vs. rhizomes). Seven seagrasses were identified to genus on the basis of leaf and stem epidermal tissue, whereas algae and rhizomes were recorded in separate categories.

For identification of ingesta fragments, a major advantage of the microscope point technique over the microhistological technique is that the ingesta is not milled to uniform size. Identification keys and materials are less developed for aquatic plants as compared to terrestrial vegetation. The fragility of aquatic plants, the relatively primitive state of the art of microscopic identification of aquatic epidermal fragments, and the close convergence of some aquatic angiosperms necessitates the use of as large a fragment as possible for Although Vavra and Holechek (1980) showed that milling of identification. terrestrial herbivore ingesta increases the proportion of identifiable plants. it is possible that milling reduces the percentage of identifiable items in manatee ingesta samples. A technique similar to the microscope point method will maintain the ingesta fragments at their original size, thereby offering maximum epidermal surface area and morphological characteristics for identifying fragments.

At this stage a modified microscope point technique is recommended for analysis of manatee ingesta. The small fragment size of manatee ingesta requires modification of the microscope point method to enable observation of the food items with a compound microscope. Preparation of microscope slides is easier than using a tray and also allows for observation of the ingesta at high magnification. In determining sampling methodology, Channels and Morrissey (1981) are followed in using several microscope slides instead of one tray (Galt et al. 1969). Holechek and Vavra (1981) recommend following Sparks and Malechek (1968) in using 5 slides with 20 fields of view each. Five points per field (100 per slide) are recorded, giving a total of 500 microscope points per sample. A minimum of 400 points per sample as recommended by Galt et al. (1968) would require 4 points recorded per field of view in this procedure.

A step-by-step outline of the method proposed here for analyzing manatee digestive tract contents follows:

- 1. Empty contents of specimen jar into enamel tray. Pick out the largest plant fragments for identification under the microscope using the photo catalogue and key.
- 2. Return remaining ingesta and preservative fluid to jar. Thoroughly mix contents with spatula. Remove 10 subsamples from jar with broad-headed forceps (each "grab" of ingesta by the forceps equals one subsample). Combine the 10 subsamples into one mass and wash under running water over a 30 mesh screen to remove dirt, diatoms, and other nuisance particles.
- 3. Remove a small subsample from the washed ingesta and spread it evenly over a 3" x 2" slide. Add 70% alcohol and cover material with cover slip. Repeat to make a total of five prepared slides from the washed ingesta. Return remaining ingesta from screen to original specimen jar.

- 4. Place slide on microscope stage and scan at 40% to 200%. Identify species present to assist later identification of fragments. If plant material is too dark or dense to observe epidermal features, remove cover slip and add Hertwig's solution to ingesta on slide, then heat over an alcohol flame until liquid boils. Remove slide from flame and add 70% alcohol to cleared ingesta and replace cover slip.
- Using a graduated mechanical stage, move slide to predetermined 5. coordinate (see Figure B-1) that defines the first field of view. View at 100X power and identify plant fragment intercepted by each line intersection of the eyepiece micrometer grid. If more than one fragment is intercepted by a line intersection, record only the fragment closest to the eye. Make five identifications (at five intersections) for each field of view, following the order illustrated An identification is recorded only for food items in Figure B-2. (i.e., do not count intersections covering sand, parasites, intestinal Continue recording grid epithelium, or other nonfood items). intersections according to the illustrated sequence in Figure B-2 until five intersections (= points = "hits") are found covering plant fragments. Identify plant fragments to species if possible. If identification of a fragment is not possible, record "unknown" for that point or hit. Record "rhizome" for points covering roots, stem, or other nonleaf material.
- 6. Move slide to the next field of view (V2) when five points have been "hit" in V1 (when five intersections have been recorded intercepting plant fragments). If the entire grid sequence (rows A-K, columns 1-11) has not yielded five hits, move the mechanical stage 1mm in the direction of the next field's coordinates (Figure B-1) and continue until five hits are achieved.

Example: If the first field (V1), at coordinate 55/125, does not cover enough material for five hits, then move the stage 1 mm toward the next field (V2 = 45/125) to coordinate 54/125. Retain recorded data from V1 and accumulate additional data from the new field (V1a) until V1's data sheet registers five total hits.

- 7. Move the stage to the coordinates of the next field. Repeat until 20 fields (V1-V20) have been recorded, with 5 hits per field, for each slide. Use a separate data sheet for each slide. Each ingesta sample will therefore have 5 data sheets, representing 5 prepared slides of 20 fields of view each, with 5 data points per field, giving a total of 500 data points per ingesta sample. Return ingesta to original sample jar when finished.
- 8. Tally data from the five data sheets onto a diet summary sheet (Figure B-3). This sheet will be a summarized record of the species encountered in the sample examined.

Because quantification of manatee diet is in its infancy, no attempt to relate frequency data to volume or weight has been attempted here. According to Vavra et al. (1978), the actual percentages of a species in the diet are probably less important than the relative proportion of that species, since the relative value ranking should identify the most important food items and thus satisfy the objectives of most diet studies. Because the method proposed above quantifies ingesta by frequency of occurrence of species identified, it will be easy to rank the individual species from the most common to the least common in the diet of manatees.

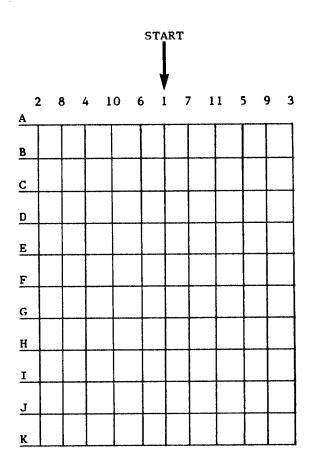


Figure B-1. Transect sequence on eyepiece grid of microscope. The transect sequence begins at column 1, row A, and proceeds down the column (1A, 1B, 1C, etc.) until five "hits" are registered. If five species have not been recorded when intersection 1K is reached, continue sequence in column 2, proceeding down (2A, 2B, 2C, etc.) until five hits are achieved. Columns 3 through 11 are likewise covered as needed, each one starting at row A, in the numerical order shown, until a total of five hits is achieved for that field.

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Figure B-2. Data sheet used to record species observed in each sample slide.

DIET SUMMARY

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SLIDE #2												100	
SLIDE #3												100	
SLIDE #4												100	
SLIDE #5												100	
TOTAL POINTS												500	
% OCCURRENCE		<u> </u>	<u> </u>									100%	
COMMENTS												· · · · · · · · · · · · · · · · · · ·	

Figure B-3. Summary data sheet used to record species observed and percent occurrence in all five sample slides.

APPENDIX C. PROVISIONAL KEY TO POTENTIAL MANATEE FOODS BASED ON MICROSCOPIC FEATURES

1.	Α.	Fragment with obvious, often large, parallel veins that glow under polarized light, and sinuous (wavy) cell walls or silica bodiesGrasses45
	В.	Fragment filamentous, terete (cylindrical), without epidermal cells, and less than 0.5 mm diameter2
	С.	Fragment terete, without epidermal cells. Surface pitted, often giving appearance of longitudinal striations. Branching stems and large spines often present at nodes. Segment width 0.30-0.60 mm
	D.	Fragment with idioblasts6
	Ε.	Fragment with obvious crystals14
	F.	Trichomes present22
	G.	Vascular tissue obvious36
	Н.	Leaf margins with short (<0.15 mm long), tooth or needle-like spines7
	I.	Epidermal cells round to oblong31
	J.	Epidermal cells sinuous anticlinal35
	Κ.	Epidermal cells regular, arranged in rows like bricks43
	L.	Epidermal cells lobed, irregular; length (0.03-0.12 mm) at least 3X cell width (0.005-0.030 mm). Fragment terete. Tips of branches with several short "spines" arranged in a spiral. Segments may glow faintly under polarized light
	M.	Epidermal cells small, more angular. Cell length less than 3X cell width21

- 2. A. Filaments branched......3
 - B. Filaments unbranched.....4
- 3. A. Filaments large, unicellular, brown, with accordion-shaped joints......Unidentified hydroid
 - B. Filaments large, with numerous attached capsules in regular series; clear to brown-colored, glowing under polarized light..........Bugala neritina

 - D. Filaments root-like, segmented, with numerous, minute multicellular "roothairs" extending from surface. Segments longer than wide, 0.30-0.50 mm long, 0.08-0.15 mm wide.........Jania adherens
 - E. Filaments segmented, with each segment composed of four long cylindrical cells. Segments joined end-to-end in a cylindrical formation. Segments 0.10-0.20 mm long, 0.04-0.07 mm wide........Polysiphonia subtilissima
- 4. A. Unbranched filaments with obvious segments......5
 - B. Unbranched filaments without obvious segments (segments minute and difficult to see); hair-like, 0.005-0.010 mm diameter0scillatoria tenuis
- 5. A. Filament segments 0.10-0.15 mm long, 0.005-0.050 mm wideOedogonium sp.
 - B. Filament segments 0.24-0.50 mm long, 0.03-0.14 mm wideSpirogyra sp./Chaetomorpha brachygona
- 6. A. "Idioblasts" (spicules) long, needle-like, non-glowing under polarized light.........Unidentified sponge
 - B. Idioblasts distinctly H-shaped, large, glowing under polarized light; druse crystals abundant in tissue. Angular to lobed epidermal cells, 0.010-0.035 mm long, 0.005-0.020 mm wide. Stomata 0.03-0.04 mm long, 0.020-0.025 mm wide. Tannin cells observed.........Rhizophora mangle

- C. Idioblasts irregularly stellate, concentrated along major vascular bundles, and glowing brightly under polarized light. Idioblasts sometimes covered with minute crystals. Vein-glow variable under polarized light. Epidermal cells round, approximately 0.02-0.05 mm diameter. Fragment flat...........Nuphar luteum
- D. Idioblasts irregularly stellate, numerous throughout fragment and glowing brightly under polarized light. Idioblasts sometimes covered with minute crystals. Vein-glow variable under polarized light. Epidermal cells round, approximately 0.02-0.05 mm diameter. Leaf edge cells often longer than other epidermal cells. Fragment flat........Nymphaea mexicana
- E. "Idioblasts" slender, coiled and worm-like; resembling "Velcro." Fragment flat..........Caulerpa prolifera
- 7. A. Spines on leaf edge only, or if on surface, only along midrib8
 - B. Spines on leaf surface and edge, unicellular.....9
- 8. A. Leaf edge spines needle-like, intermittent......10
 - B. Leaf edge spines variable in shape; leaf edge cells same length or shorter than other epidermal cells, tannin bodies present............11
 - C. Leaf edge spines tooth-like; leaf edge cells often longer than other epidermal cells......12
- 10. A. Spines on one margin of leaf only, not abundant. Leaf tip distinct, with two opposing spines. Epidermal cells mostly angular, 0.02-0.05 mm long, 0.02-0.03 mm wide. Small unidentified crystals observed throughout fragment. Tannin cells observed.........Ceratophyllum demersum

- 11. A. Leaf edge spines robust, common. Tannin bodies globular to filamentous. Epidermal cells block to hexagon-shaped, large (0.03-0.06 mm long, 0.02-0.05 mm wide), arranged in rows. Leaf blade broad, >3.0 mm wide. Numerous parallel veins obvious, and usually glowing under polarized light; cross veins not obvious. Globular trichomes rare. Stomata and crystals absent..........Vallisneria americana
 - B. Leaf edge spines short and irregularly spaced, restricted mostly to tip of leaf. Tannin bodies common, oval to rectangular, evenly distributed on leaf surface. Leaf blade narrow, <3.0 mm wide, flat, unbranched. Veins not obvious. Epidermal cells mostly angular, small, 0.01-0.03 mm long, 0.01-0.02 mm wide, arranged in rows. Secretory cells (possibly empty tannin sacs) common. Mesophyll not apparent. Leaf tip acute, with small teeth...........Ruppia maritima
- 12. A. Upper epidermal cells considerably larger than lower epidermal cells...........13
 - B. Epidermal cells on both surfaces of leaf approximately the same size, 0.04-0.12 mm long, 0.02-0.05 mm wide, angular, arranged in rows. Leaf edge veins glow brightly under polarized light. Leaf tip with one or two spines...........Najas guadalupensis
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 - D. Small crystal sand irregularly abundant along or above veins. Short to long macrohairs common over epidermis. Multicellular scales (peltate hairs) small and numerous. Vascular tissue abundant, reticulate. Veins glow brightly under polarized light. Epidermal cells angular to sinuous anticlinal, 0.015-0.090 mm long, 0.01-0.04 mm wide. Stomata 0.015-0.030 mm long, 0.010-0.015 mm wide.......Fraxinus caroliniana
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 - <u>Note:</u> Our limited microscopic observations on *Pontederia* suggest *Eichhornia crassipes* and *Pontederia cordata lancifolia* have nearly identical microhistological features. This key does not distinguish between the two species.

- 19. A. Raphides and druse crystals numerous. Venation obvious, reticulate. Epidermal cells angular, 0.010-0.065 mm long, often with a single papilla centered on each cell.........Colocasia esculenta
 - B. Crystal sand and styloids also present. Vascular tissue abundant, reticulate, midrib prominent. Epidermal cells irregularly lobed to weakly sinuous anticlinal, 0.02-0.16 mm long, 0.02-0.05 mm wide. Leaf edge cells often longer than other epidermal cells......Ludwigia repens
 - C. Epidermal cells lobed to weakly sinuous anticlinal, 0.02-0.05 mm long, 0.02-0.03 mm wide. Leaf edge cells often longer than other epidermal cells. "Leaflets" (fronds) minute, less than 4.0 mm long, with reticulate venation......Lemna sp.
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- 20. A. Raphides common. Crystal sand variable in abundance, small. Tannin bodies numerous, small and globular. Numerous parallel veins present that glow (often multicolored) under polarized light. Epidermal cells angular to lobed, 0.01-0.03 mm long. Five or more rows of stomata between veins. Stomata 0.015-0.030 mm long, 0.010-0.015 mm wide......Typha domingensis
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- 29. A. Long, unicellular macrohairs abundant, multicellular club-hairs common. Stomata 0.02-0.03 mm long, 0.010-0.015 mm wide. Large crystal sand common. Angular epidermal cells 0.01-0.05 mm long, 0.01-0.03 mm wide, covered with papillae. Veins robust, reticulate..........Diospyros virginiana
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- 30. A. Deciduous conical spines on leaflet surface and edge, often evident only by their disk-shaped bases. Epidermal cells angular to lobed, 0.04-0.20 mm long, 0.02-0.04 mm wide. Leaf edge cells often longer than other epidermal cells. Leaflets 0.50-0.90 mm wide. Leaf tips with two spines. No crystals present. Tannin cells observed..........Cabomba caroliniana
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- B. Leaf blade flat, unbranched. Broad midrib more obvious than leaf edge veins. Cross veins sometimes apparent. Veins do not glow under polarized light. Epidermal cells arranged in rows like bricks, block-shaped, often hexagonal, 0.02-0.03 mm long, 0.01-0.02 mm wide. Leaf tip pointed, without spines. Mesophyll apparentZannichellia palustris
- 39. A. Veins and cross veins numerous, usually glowing under polarized light. Epidermal cells lobed to angular, 0.03-0.10 mm long, 0.02-0.05 mm wide, not in linear series. Stomata 0.04-0.05 mm long, 0.02-0.03 mm wide. Unidentified crystals common...........Sagittaria lancifolia
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 - B. Epidermal cells block-shaped, often hexagonal, 0.02-0.03 mm long, 0.01-0.02 mm wide. Leaf edge cells same length as other epidermal cells. Mesophyll apparent. Veins few with broad midrib more obvious than leaf edge veins. Cross veins sometimes apparent. Veins do not glow under polarized light. Leaf tip pointed, without spines......Zannichellia palustris
- 45. A. Papillae absent......46
 - B. Papillae present......49

- 46. A. Silica bodies present......47
 - B. No silica bodies or silica/tannin cells present. Veins 0.02-0.10 mm wide, narrower than intercostal region, with six or more rows of stomata between them. Stomata numerous, 0.02-0.03 mm long, 0.015-0.025 mm wide. Spines, trichomes, and papillae absent........Juncus sp.
- 47. A. Prickles on veins common......Panicum spp.

Panicum commutatum: Veins narrower than intercostal region, 0.02-0.09 mm wide, with one row of cross or dogbone-shaped silica bodies (except for middle vein, which has three rows) over veins. Veins separated by two to three rows of stomata. Stomata 0.02-0.03 mm long, 0.010-0.025 mm wide. One row of unidentified objects (possibly prickles) between veins. Microhairs club-shaped. Spines numerous on leaf margins.

Panicum dichotomum: Veins 0.02-0.06 mm wide, narrower than intercostal region; separated by two rows of stomata. Stomata 0.030-0.045 mm long, 0.01-0.02 mm wide. Microhairs often with long, distal cell, usually the same size as basal cell. One row of unidentified objects (possibly prickles) between veins. Silica cell pairs few. Epidermal cells very sinuous. Spines numerous on leaf margins.

Panicum hemitomon: Veins 0.01-0.04 mm wide, much narrower than intercostal region, separated by one to two rows of stomata. Stomata 0.02-0.04 mm long, 0.01-0.02 mm wide. Silica bodies on veins dogbone to nodular-shaped. One row of silica bodies between veins, irregular, mostly tall and narrow, sometimes crenate. Microhairs club and hair-shaped. Spines numerous on leaf margins.

- B. Veins without prickles or prickles infrequent......48
- 48. A. Prickles infrequent; 0.020-0.025 mm long between veins and 0.04-0.08 mm long above veins. Veins 0.01-0.03 mm wide and narrower than intercostal region, separated by two rows of stomata. Stomata 0.02-0.04 mm long, 0.01-0.02 mm wide. Macrohairs abundant between veins. Microhairs club and hair-shaped. Occasional cross-shaped silica bodies between veins. Silica bodies over veins nodular and dogbone-shaped, mostly in one row (middle vein of leaf has three rows). Spines abundant and overlapping on leaf margins..........Oplismenus setarius

- B. Prickles on veins uncommon. Silica bodies large, nodular to dogbone-shaped on veins, not apparent in intercostal region. Veins 0.02-0.06 mm wide, narrower than intercostal region. Veins separated by two to four rows of stomata. Stomata 0.035-0.050 mm long, 0.015-0.030 mm wide. Stomata subsidiary cells mostly low dome-shaped, but also triangular. Paired short cells uncommon. Microhairs club-shaped. Leaf edge with small bumps (papillae?) and large spines. Crystal sand common............Sacciolepis striata
- 49. A. Most papillae small, dot-like......50
 - B. Papillae not as above......52
- 50. A. Minute papillae abundant, all of equal size........51
 - B. Minute papillae associated with much larger, dome-shaped papillae. Leaf margins bordered with large hook-shaped spines and numerous papillae. Prickles between and above veins. Large prickles over veins abundant. Veins 0.03-0.06 mm wide, narrower than intercostal region. Main veins separated by two to four rows of stomata. Stomata 0.01-0.04 mm long, 0.005-0.020 mm wide. Dogbone-shaped silica bodies present at right angles to long axes of veins and appearing as two rows of oval bodies between prickles. Silica bodies over veins cross-shaped from edge-view, oblong to dogbone-shaped from above. Microhairs very small.......Leersia hexandra

- 52. A. Epidermal cells with only one papilla......53
 - B. Epidermal cells with two or more papillae.....54
- - B. One papilla per long cell, wider than tall and appearing as a shallow, wave-like bump. Veins 0.01-0.08 mm wide, usually narrower than intercostal region. Microhairs rare......Echinochloa spp.

Echinochloa muricata: Veins with one to three rows of silica bodies, dogbone or nodular-shaped over veins, with middle row dogbone-shaped and outer rows tall and narrow, or block to cross-shaped between veins. Prickles present over and between veins. One to three (usually two) rows of stomata between veins, 0.025-0.030 mm long, 0.02 mm wide.

Echinochloa paludigena: Veins with one to three rows of silica bodies, dogbone or nodular-shaped over veins, dogbone-shaped between veins. Prickles present over and between veins. Two rows of stomata between veins, 0.025-0.040 mm long, 0.01-0.02 mm wide.

- - B. Papillae small to medium in height, conical to wave-like. Papillae obscure all other epidermal features. Veins deep, broad, 0.02-0.07 mm wide, narrower than intercostal region. Large prickles over and between veins. Silica bodies abundant on veins, not in rows but scattered throughout. Two rows of stomata between veins. Stomata 0.015-0.020 mm long, 0.01 mm wide........Distichlis spicata

GLOSSARY

cork cell a short cell with waxy, thickened walls; characteristic of

the Gramineae epidermis.

costal the epidermal region over a leaf vein.

crenate having a notched or scalloped margin.

cross vein a strand of leaf vascular tissue connecting one major vein

element to another.

crystals in plants, inorganic deposits of calcium oxalate or silica.

crystal sand minute, prismatic crystals.

culm the stem of grasses and sedges.

cuticle a wax-like layer of the outer portions of the epidermal

cells.

druse a compound crystal, spherical in shape, with many protruding

component crystals.

epidermis the outermost cell layer of the plant.

guard cells a pair of specialized epidermal cells surrounding the

stomatal opening.

hair in plants, a trichome; usually straight and slightly tapered

from base to tip.

intercostal the epidermal region between leaf veins.

idioblast a specific cell clearly distinguished from the other tissue

cells by size, structure or content (see Nuphar, Nymphaea,

and Rhizophora illustrations for examples).

interstomal in the Gramineae and allies, the epidermal region between

aligned stomata.

macrohair a relatively long, thick-walled trichome usually, but not

always, unicellular.

mesophyll the parenchymatous tissue between the two epidermal layers of the leaf.

microhair a uni- or multicellular microscopic trichome. Commonly bicellular, with the basal cell relatively thick-walled and apical cell thin-walled and delicate.

papillae minute, nipple-shaped projections of a plant's epidermal cuticle.

peltate mushroom-shaped; as with some trichomes.

phloem the tissue that transports food and minerals in vascular plants; characterized by sieve elements, parenchyma cells, fibers and sclereids.

prickle a short and pointed hair, often thorn-shaped. (Also referred to in the literature as: asperity, bristle, emergence, or hook.)

raphide needle-shaped crystals occurring in a dense bundle, often enclosed in a mucilage-filled cell.

reticulate a net-like arrangement of vascular tissue.

secretory cells specialized cells that produce a variety of secretions on the surface of the plant.

silica body an amorphous body of silica in the epidermal cells of the Gramineae.

a short epidermal cell of grass leaf and culm that often contains one silica body of various shapes.

sinuous anticlinal having a margin with many wavy indentations in all directions. (See epidermal cell shapes illustrated on page 19).

spicule skeletal elements of a sponge; of various sizes and shapes but often stylate.

spine a thick-walled, short and stout hair-like projection on the edge (usually) of a leaf; of a variety of shapes and sizes.

stellate star-shaped; radiating.

a minute opening, with guard cells, in the epidermis of plants.

stomata plural of stoma.

stomate with stoma or stomata.

styloid

a thick, elongated crystal.

subsidiary cell

an epidermal cell that borders on a guard cell of the stoma and which differs from the ordinary epidermal cells.

tannin

a heterogenous group of phenolic compounds (usually brown in color) that may form droplets in the cytoplasm, fill entire cells, penetrate cell walls or be found in idioblasts.

tannin bodies

fused droplets of tannin in specialized containers (tannin sacs) or the cytoplasm proper.

tannin cell

a specialized secretory cell containing tannin.

terete

cylindrical.

tracheary element

one of either type (tracheid and vessel member) of xylem element that takes part in water transport.

trichome

a hairlike outgrowth of a plant's epidermis; of various shapes, sizes, structures, and functions. Includes hairs, scales, papillae, spines, and glands.

vascular tissue

the tissue of plants containing xylem, phloem, or both.

vein

a strand of vascular tissue.

xylem

the water-conducting tissue of vascular plant characterized by the presence of tracheary elements.

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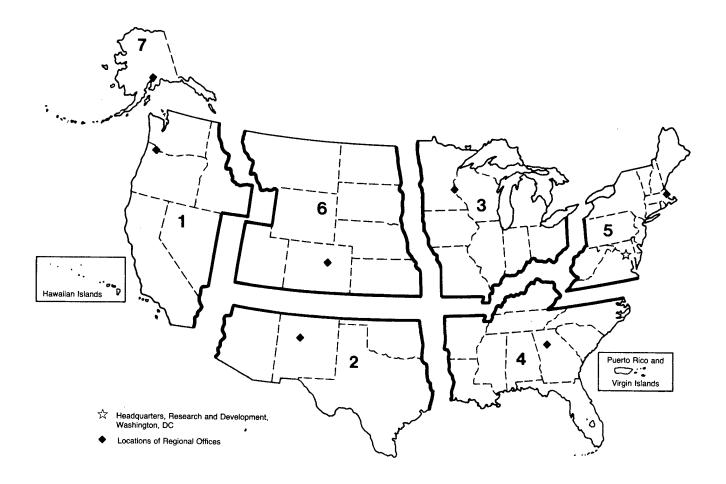
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a reference slide collection is described. Techniques for food identification and quantification for 83 plant species and three invertebrate groups are presented to provide a standardized method for the study of the manatee diet in Florida. Methods used for photomicrography of plant material and preparation of ingesta and fecal material for analysis are included. A diagnostic key of microhistological characters, summary tables of distinct features, and character matrix tables are included to aid in the identification of unknown plant fragments recovered from manatee ingesta and fecal samples. 17. Document Analysis a. Descriptors	
b. Identifiers/Open-Ended Terms	
aquatic plants manatee diet <u>Trichechus</u> <u>manatus</u> Florida	
c. COSATi Field/Group	
18. Availability Statement	19. Security Class (This Report) 21. No. of Pages
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